

MULTISPECIFIC ANTIBODIES HAVING SPECIFICITY FOR TNF α AND IL-17A, ANTIBODIES TARGETING IL-17A, AND METHODS OF USE THEREOF

FIELD OF THE INVENTION

The present invention relates to an isolated multispecific antibody comprising a first
5 domain specifically binding TNF α and a second domain specifically binding IL-17A and,
optionally, a third domain specifically binding human serum albumin. The present invention
further relates to methods of use of said antibody, pharmaceutical compositions and methods
of use thereof, and a kit comprising said antibody. The present invention also relates to a
nucleic acid comprising a nucleotide sequence encoding said antibody, a vector comprising
10 said nucleic acid, a host cell comprising said nucleic acid or said vector, and a method of
producing said antibody.

The present invention furthermore relates to an isolated antibody which specifically binds
human IL-17A, a multispecific molecule comprising said isolated antibody of the invention,
pharmaceutical compositions and methods of use thereof. The present invention further
15 relates to a kit comprising said antibody, a nucleic acid comprising a nucleotide sequence
encoding said antibody, a vector comprising said nucleic acid, a host cell comprising said
nucleic acid or said vector, and a method of producing said antibody.

BACKGROUND OF THE INVENTION

TNF α is a homo-trimeric pro-inflammatory cytokine that is released by and interacts with
20 cells of the immune system. TNF α is found as a soluble protein as well as a precursor form
called transmembrane TNF α that is expressed as a cell surface type II polypeptide.
Transmembrane TNF α is processed by metalloproteinases such as TNF α -converting enzyme
(TACE) between residues Ala76 and Val77, resulting in the release of the soluble form of
TNF α of 157 amino acid residues. Soluble TNF α is a homotrimer of 17-kDa cleaved
25 monomers. Transmembrane TNF α also exists as a homotrimer of 26-kD uncleaved
monomers. Both membrane and soluble TNF α are biologically active. TNF α can bind to two
receptors, TNF receptor 1 (TNFR1) and 2 (TNFR2), wherein the transmembrane TNF α acts
predominantly via TNFR2. TNFR1 is widely expressed on a variety of cells and its
engagement triggers pro-inflammatory responses. TNFR2 expression is almost exclusively

restricted to immune cells and its binding promotes cell survival and proliferation (Bazzoni F, Beutler B, N Engl J Med (1996) 334(26):1717–25; Locksley RM, et al., Cell (2001) 104(4):487–501; Cabal-Hierro L, Lazo PS, Cell Signal (2012) 24(6):1297–305; Brenner D, et al., Nat Rev Immunol (2015) 15(6):362–74).

5 TNF α has been shown to be up-regulated in a number of human diseases, such as inflammatory and autoimmune disorders, including chronic diseases such as rheumatoid arthritis, Crohn's disease, ulcerative colitis and multiple sclerosis. Antibodies to TNF α have been proposed for the prophylaxis and treatment of endotoxic shock (Beutler et al., Science, 234, 470-474, 1985). Bodmer et al., (Critical Care Medicine, 21, S441-S446, 1993) and
10 Wherry et al., (Critical Care Medicine, 21, S436-S440, 1993) discuss the therapeutic potential of anti-TNF α antibodies in the treatment of septic shock. The use of anti-TNF α antibodies in the treatment of septic shock is also discussed by Kirschenbaum et al., (Critical Care Medicine, 26, 1625-1626, 1998). Collagen-induced arthritis can be treated effectively using an anti-TNF α monoclonal antibody (Williams et al. (Proc. Natl. Acad. Sci. U.S.A. 89, 9784-
15 9788, 1992)). The use of anti-TNF α antibodies in the treatment of rheumatoid arthritis and Crohn's disease is discussed in Feldman et al. (Transplantation Proceedings, 30, 4126-4127, 1998), Adorini et al. (Trends in Immunology Today, 18, 209-211, 1997) and in Feldman et al. (Advances in Immunology, 64, 283-350, 1997). The antibodies to TNF α previously used in such treatments are generally chimeric antibodies, such as those described in U.S. Pat. No.
20 5,919,452.

Monoclonal antibodies against TNF α have been described in the prior art. Meager et al. (Hybridoma, 6, 305-311, 1987) describe murine monoclonal antibodies against recombinant TNF α . Fendly et al. (Hybridoma, 6, 359-370, 1987) describe the use of murine monoclonal antibodies against recombinant TNF α in defining neutralizing epitopes on TNF α .
25 Furthermore, in International Patent Application WO 92/11383, recombinant antibodies, including CDR-grafted antibodies, specific for TNF α are disclosed. Rankin et al. (British J. Rheumatology, 34, 334-342, 1995) describe the use of such CDR-grafted antibodies in the treatment of rheumatoid arthritis. U.S. Pat No. 5,919,452 discloses anti-TNF α chimeric antibodies and their use in treating pathologies associated with the presence of TNF α . Further
30 anti-TNF α antibodies are disclosed in Stephens et al. (Immunology, 85, 668-674, 1995), GB-A-2 246 570, GB-A-2 297 145, US 8,673,310, US 2014/0193400, EP 2 390 267 B1, US 8,293,235, US 8,697,074, WO 2009/155723 A2 and WO 2006/131013 A2.

Currently approved anti-TNF α biotherapeutics include (i) infliximab, a chimeric IgG anti-human monoclonal antibody (Remicade®; Wiekowski M et al: “Infliximab (Remicade)”, Handbook of Therapeutic Antibodies, WILEY-VCH; Weinheim, 2007-01-01, p.885-904); (ii) etanercept, a TNFR2 dimeric fusion protein, with an IgG1 Fc (Enbrel®); (iii) adalimumab, a
5 fully human monoclonal antibody (mAb) (Humira®; Kupper H et al: “Adalimumab (Humira)”, Handbook of Therapeutic Antibodies, WILEY-VCH; Weinheim, 2007-01-01, p.697-732); (iv) certolizumab, a PEGylated Fab fragment (Cimzia®; Melmed G Y et al: “Certolizumab pegol”, Nature Reviews. Drug Discovery, Nature Publishing Group, GB, Vol. 7, No. 8, 2008-08-01, p.641-642); (v) Golimumab, a human IgG1K monoclonal antibody
10 (Simponi®; Mazumdar S et al: “Golimumab”, mAbs, Landes Bioscience, US, Vol. 1, No. 5, 2009-09-01, p.422-431).

However, anti-TNF α treatment was shown to have certain limitations. Not all patients achieve sufficient clinical response or maintain clinical response to anti-TNF α over time, resulting in a need to switch to a new therapy to control their disease. For example upon anti-
15 TNF α treatment of rheumatoid arthritis (RA) patients, approximately 40% of patients never respond, and only 20% of patients experience a major reduction in disease activity. Hence, there continues to be a considerable unmet clinical need for treatments regarding more effective suppression of progression of such disorders as inflammation and autoimmune disorders.

20 Activation of a complementary biological pathway in patients after anti-TNF α treatment, may be one of the reasons why many patients fail to respond to the anti-TNF α therapy or have only partial response. Recently emerged body of evidence indicates on the role of IL-17A in the pathogenesis of inflammatory and autoimmune disorders, for example in RA.

25 The interleukin 17 (IL-17) family in human and in mice is composed of six cytokines, IL-17A, IL-17B, IL-17C, IL-17D, IL-17E (also called IL-25) and IL-17F, and plays roles in acute and chronic inflammatory responses. The interleukin 17 receptor (IL-17R) family consists of five members, namely IL-17RA, IL-17RB, IL-17RC, IL-17RD and IL-17RE.

Interleukin-17A (IL-17A or IL17A, synonymous with IL-17 or cytotoxic T-lymphocyte-associated antigen-8 (CTLA-8)) is a homodimeric pro-inflammatory cytokine. IL-17A is
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produced by a subset of memory CD4⁺ T cells (termed Th17), CD8⁺ T cells (Tc17), invariant NKT cells, $\gamma\delta$ T cells, non-T non-B lymphocytes (termed type 3 innate lymphoid cells) and neutrophils. IL-17A and IL-17F form a distinct subgroup within the IL-17 family. They share the greatest sequence homology and identity within IL-17 family, while other members of the

5 IL-17 family have a significantly lower sequence identity to IL-17A (Starnes, T., et al., J Immunol. 167(8):4137-40 (2001); Aggarwal, S. and Gurney, A. L., J. Leukoc Biol, 71(1): 1-8 (2002)). Both IL-17A and IL-17F signal through a heterodimeric receptor complex composed of IL-17RA and IL-17RC (Toy D., et al., J Immunol. 2006; Wright JF., et al., J Immunol. 2008; 181(4):2799–2805). IL-17A and IL-17F can form IL-17A/A or IL-17F/F disulphide

10 linked homodimers and IL-17A/F disulphide linked heterodimers (Wright JF. et al., J Immunol. 2008; 181(4):2799–2805; Liang SC. et al., J Immunol. 2007; 179(11):7791–7799). IL-17A as well as IL-17F induces expression of pro-inflammatory cytokines and anti-microbial peptides.

Human IL-17A (CTLA-8, Swiss Prot Q 16552, also referred to as IL-17 or IL17; SEQ ID

15 NO: 33) is implicated in various inflammatory conditions such as autoimmune diseases, metabolic disorders and cancer (Ouyang W., et al., Immunity. 2008; 28(4):454–467; Milner JD., Curr Opin Immunol. 2011; 23(6):784–788; Kuchroo VK., et al., Nat Med. 2012; 18(1):42–47; Ahmed M. and Gaffen SL., Cytokine Growth Factor Rev. 2010; 21(6):449–453; Trinchieri G., Annu Rev Immunol. 2012; 30:677–706; Gallimore AM, Godkin A., N Engl J

20 Med. 2013; 368(3):282–284; Ye P., et al., J Exp Med. 2001; 194(4):519–527; Chung DR., et al., J Immunol. 2003; 170(4):1958–1963; Huang W., et al., J Infect Dis. 2004; 190(3):624–631; Ishigame H., et al., Immunity. 2009; 30(1):108–119; see for review Gu C., et al., Cytokine. 2013 Nov 64(2)). IL-17A plays a role in the induction of other inflammatory

25 cytokines and chemokines for neutrophil recruitment, acute phase proteins, anti-microbial peptides, mucins, matrix metalloproteinases and adhesion molecules. IL-17A also synergizes with other cytokines including TNF α and IL-1 beta to further induce chemokine expression (Chabaud M., et al., J. Immunol. 161(1):409-14 (1998)).

Pathological production of IL-17A leads to excessive inflammation and tissue damage (see for review Gu et al., Cytokine. 2013 November; 64(2)). High IL-17A levels were found

30 in multiple sclerosis (MS), psoriasis, asthma, Crohn's disease and rheumatoid arthritis patients. Treatment of animals with IL-17A neutralizing antibodies decreases disease incidence and severity in autoimmune encephalomyelitis (Komiyama, Y. et al, J. Immunol.

177 (2006) 566-573). In addition, IL-17A neutralizing antibodies reduce severity and incidence of mouse rheumatoid arthritis model of collagen induced arthritis, and high levels of IL-17A can be detected in the synovial fluid of inflamed joints from rheumatoid arthritis patients (Ziolkowska, M. et al, J. Immunol. 164 (2000) 2832-2838; Kotake, S. et al, J. Clin. Invest. 103 (1999) 1345-1352; Hellings P.W. et al, Am. J. Resp. Cell Mol. Biol. 28 (2003) 42-50).

A large body of experimental evidence in human and animal models has supported the development of IL-17A-targeted therapies. Several anti-IL-17 antibodies were developed including AIN457 (secukinumab; see U.S. Patent No.7, 807,155 and WO 2006/013107), LY2439821 (ixekizumab; see U.S. Patent Nos. 7,838,638 and 8,110,191 and WO 2007/070750), SCH900117 (Merck), RG4943 (Roche), etc. Examples of anti-IL-17A antibodies are disclosed in WO 2006/013107, WO 2006/054059, WO 2007/070750, WO 2007/149032, WO 2008/001063, WO 2008/021156, WO 2010/034443, WO 2010/102251, WO 2012/018767, WO 2014/161570, WO 2014/001368, WO 2014/122613, WO 2015/070697, WO 2015/137843, WO 2016/048188, WO 2016/113557, WO 2016/138842, WO 2017/068472.

Several clinical trials with various molecules blocking IL-17A signaling have been conducted or are still ongoing. The biologics targeting either IL-17A or its receptor and their efficacies are being evaluated in the setting of inflammatory or autoimmune disorders, such as rheumatoid arthritis, ankylosing spondyloarthritis, Crohn's disease, psoriasis, multiple sclerosis and ozone-induced neutrophilia.

For example, secukinumab, a fully human IgG1 κ anti-IL-17A monoclonal antibody (U.S. Patent No.7, 807,155 and WO 2006/013107), is now approved for the treatment of psoriasis, psoriatic arthritis and ankylosing spondylitis behavior (see for review Wang et al., Eur J Rheumatol 2017 (4) 272-7). In phase III studies assessing the long-term efficacy and safety of secukinumab in subjects with psoriatic arthritis (FUTURE I and FUTURE II), secukinumab was significantly more effective than placebo in improving the signs and symptoms of psoriatic arthritis (Mease PJ. et al. N Engl J Med 2015; 373: 1329-39; McInnes IB. et al. The Lancet; 386:1137-46).

Ixekizumab, a humanized anti-IL-17A monoclonal antibody (U.S. Patent Nos. 7,838,638 and 8,110,191 and WO 2007/070750), was studied in biologic-naïve patients with active psoriatic arthritis in a 24-week phase III trial (SPIRIT-P1) (Mease PJ. Et al., Ann Rheum Dis. 2017 Jan;76(1):79-87). It was demonstrated that in biologic-naïve patients with active
5 psoriatic arthritis, ixekizumab treatment resulted in improvements in disease activity and physical function, as well as in the inhibition of structural damage progression. Ixekizumab was also shown to be effective in treating patients with moderate-to-severe plaque psoriasis (Griffiths CEM, et al., The Lancet; 386: 541-51).

Brodalumab is a fully human IL-17 receptor (IL-17RA) monoclonal antibody (see U.S.
10 Patent No. 7,767,206), and has been proven effective in the treatment of psoriasis (Papp KA. et al. N Engl J Med 2012; 366: 1181-9). It also showed significant and sustained response in psoriatic arthritis patients in a placebo-controlled phase II study (Mease PJ. et al., N Engl J Med 2014; 370: 2295-306). However, treatment with brodalumab has been coupled to strong adverse events, such as upper respiratory tract infection, fatigue, diarrhea, and reported
15 suicidal thoughts and behavior (see for review Wang et al., Eur J Rheumatol 2017 (4) 272-7).

Accordingly, IL-17A is a promising target in the therapy of inflammatory and autoimmune disorders. Although a number of anti-IL-17A antibodies have been identified up to date, there is still a need for the development of improved therapeutic antibodies being able to effectively reduce or eliminate IL-17A activity in inflammatory responses and autoimmune
20 diseases, and at the same time having improved safety profile and being suitable for development. The therapeutic antibodies should have, in addition to beneficial affinity, efficacy and immunogenicity, improved biophysical properties leading to better developability, producibility in high yields and protein stability. A treatment whereby both the TNF α and IL-17A biological pathways are simultaneously blocked, has the potential to
25 significantly improve response rates and address the unmet need in the treatment of disorders mediated by TNF α and IL-17A. Several biologic therapeutics that specifically bind IL-17 and TNF α have been proposed up to date. WO 2010/102251 (Abbvie Inc.) discloses dual-specific tetravalent antibodies which bind to both TNF α and IL-17. WO 2013/063110 (Abbvie Inc.) discloses a multivalent DVD-Ig binding protein capable of binding TNF and IL-17. WO
30 2014/044758 (Covagen ACJ) discloses a fusion construct capable of inhibiting glycosylated IL-17A and binding TNF α . Also, WO 2014/137961 (Eli Lilly and Company) and WO 2017/132457 (Janssen Biotech) disclose anti-TNF and anti-IL-17A bispecific antibodies. WO

2017/102830 (UCB Biopharma) discloses a multispecific antibody, which is capable of inhibiting TNF α , IL-17A and IL-17F, in particular, comprising a binding domain specific to human TNF α and a binding domain specific to human IL-17A and human IL-17F. Xu et al., Oncotarget, 8 (2017) 81860-81872, generated an IgG-like bispecific antibody (bsAb), wherein
5 the two arms of the IgG-like molecules are directed against TNF α and IL-17A, respectively. Interestingly, WO 2015/014979 (Roche, see also Fischer et al., Arthritis & Rheumatology 67 (2015) 51–62) discloses bispecific tetravalent IL-17AxTNF antibody constructs, which are either bivalent (“2 +2” construct) or monovalent (“2 +2” construct) for each of the two antigens. According to WO 2015/014979, the bivalent construct is to be preferred over the
10 monovalent alternative.

However, there still remains a need for improved anti-inflammatory drugs that can effectively neutralize the activity of both IL-17A and TNF α for the treatment of disorders such as inflammatory and autoimmune disease, for example rheumatoid arthritis, in which a significant portion of patients still do not respond adequately to therapy. There is still a need
15 for the development of improved therapeutic antibodies effectively neutralizing the activity of both IL-17A and TNF α , having beneficial affinity and efficacy, and improved safety profile, e.g., lower immunogenicity. Furthermore, the therapeutic antibodies should have improved biophysical properties leading to better developability, producibility in high yields and superior antibody stability.

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SUMMARY OF THE INVENTION

It is an object of the present invention to provide a medicament to improve treatment of inflammatory and autoimmune disorders.

The antibody of the invention provides a new treatment option for patients with an
25 unmet medical need. The present invention provides a novel multispecific antibody, which is capable of simultaneously inhibiting IL-17A and TNF α , and which has further improved properties beneficial for use in therapies, such as higher affinity, improved efficacy, selectivity, safety, e.g., low immunogenicity, and improved biophysical properties, such as developability, and stability.

In one aspect, the present disclosure relates to isolated multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , and, optionally, a third domain specifically binding human serum albumin.

5 In one aspect, the present disclosure relates to a pharmaceutical composition comprising the multispecific antibody of the invention and a pharmaceutically acceptable carrier.

In a further aspect, the present disclosure provides the multispecific antibody of the invention or the pharmaceutical composition of the invention for use as a medicament.

10 In a further aspect, the present disclosure provides the multispecific antibody of the invention or the pharmaceutical composition of the invention for use in treatment of a disorder mediated by IL-17A and/or TNF α or a disorder that can be treated by inhibiting Gro- α secretion, in particular for use in the treatment of an inflammatory condition or an autoimmune disease.

15 In one aspect, the present disclosure provides use of the multispecific antibody of the invention or the pharmaceutical composition of the invention for the treatment of a disorder mediated by IL-17A and/or TNF α or a disorder that can be treated by inhibiting Gro- α secretion, in particular for use in the treatment of an inflammatory condition or an autoimmune disease.

20 In one aspect, the present disclosure provides use of the multispecific antibody of the invention or the pharmaceutical composition of the invention in the manufacture of a medicament for treatment of a disorder mediated by IL-17A and/or TNF α or a disorder that can be treated by inhibiting Gro- α secretion, in particular for use in the treatment of an inflammatory condition or an autoimmune disease.

25 In yet another aspect, the present disclosure provides a method of treating a disorder mediated by IL-17A and/or TNF α , said method comprising administering an effective amount of the multispecific antibody of the invention or the pharmaceutical composition of the invention to a subject in need thereof.

In one aspect, the present disclosure provides a kit comprising the multispecific antibody of the invention, or the pharmaceutical composition of the invention.

In a further aspect, the present disclosure provides a nucleic acid comprising a nucleotide sequence encoding the multispecific antibody of the invention. In a further aspect, the present disclosure provides a vector comprising said nucleic acid. In a further aspect, the present disclosure provides a host cell comprising said nucleic acid or said vector.

5 In yet another aspect, the present disclosure provides a method of producing the multispecific antibody of the invention or a binding domain thereof or a fragment thereof, the method comprising the step of culturing a host cell comprising a nucleic acid or a vector encoding the multispecific antibody of the invention or a binding domain thereof or a fragment thereof.

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The aspects, advantageous features and preferred embodiments of the present disclosure, summarized in the following items, respectively alone or in combination, further contribute to solving the object of the invention:

- 15 1. An isolated multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α .
2. The multispecific antibody of item 1, wherein said antibody comprises only one domain specifically binding IL-17A and/or only one domain specifically binding TNF α .
- 20 3. The multispecific antibody of item 1 or item 2, wherein said antibody is capable of neutralizing the biological activity of human TNF α and human IL-17A.
4. The multispecific antibody of any one of the preceding items, wherein said antibody selectively binds to human IL-17A over human IL-17B, IL-17C, IL-17D, IL-17E and IL-17F as measured by ELISA.
- 25 5. The multispecific antibody of any one of the preceding items further comprising a third domain having a specificity against an antigen other than IL-17A or TNF α .
6. The multispecific antibody of item 5, wherein said antibody comprises a third domain specifically binding to human serum albumin (HSA), preferably said antibody comprising only one domain specifically binding human serum albumin.
- 30 7. The multispecific antibody of any one of the preceding items, wherein said domains are capable of binding to their respective antigen or receptor simultaneously.
8. The multispecific antibody of any one of the preceding items, wherein said first domain and said second domain, and, optionally said third domain, are independently

- selected from the group consisting of a Fab, an Fv, an scFv, dsFv, an scAb, STAB, a single domain antibody (sdAb or dAb), a single domain heavy chain antibody, and a single domain light chain antibody, a VHH, a VNAR, single domain antibodies based on the VNAR structure from shark, and binding domains based on alternative scaffolds including but limited to ankyrin-based domains, fynomers, avimers, anticalins, fibronectins, and binding sites being built into constant regions of antibodies (e.g. F-star's Modular Antibody TechnologyTM), preferably from the group consisting of a Fab, Fv and an scFv, more preferably wherein said first domain and/or said second domain and/or said third domain is/are Fv or scFv.
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- 10 9. The multispecific antibody of any one of the preceding items, wherein said multispecific antibody is in a format selected from the group consisting of a single-chain diabody (scDb), a tandem scDb (Tandab), a linear dimeric scDb (LD-scDb), a circular dimeric scDb (CD-scDb), a bispecific T-cell engager (BiTE; tandem di-scFv), a tandem tri-scFv, a tribody (Fab-(scFv)₂) or bibody (Fab-(scFv)₁), Fab, Fab-Fv₂, Morrison (IgG CH3-scFv fusion (Morrison L) or IgG CL-scFv fusion (Morrison H)), triabody, scDb-scFv, bispecific Fab₂, di-miniantibody, tetrabody, scFv-Fc-scFv fusion, scFv-HSA-scFv fusion, di-diabody, DVD-Ig, COVD, IgG-scFab, scFab-dsscFv, Fv₂-Fc, IgG-scFv fusions, such as bsAb (scFv linked to C-terminus of light chain), Bs1Ab (scFv linked to N-terminus of light chain), Bs2Ab (scFv linked to N-terminus of heavy chain), Bs3Ab (scFv linked to C-terminus of heavy chain), Ts1Ab (scFv linked to N-terminus of both heavy chain and light chain), Ts2Ab (dsscFv linked to C-terminus of heavy chain), bispecific antibodies based on heterodimeric Fc domains, such as Knob-into-Hole antibodies (KiHs); an Fv, scFv, scDb, tandem-di-scFv, tandem tri-scFv, Fab-(scFv)₂, Fab-(scFv)₁, Fab, Fab-Fv₂, COVD fused to the N- and/or the C-terminus of either chain of a heterodimeric Fc domain or any other heterodimerization domain, a MATCH and DuoBodies, preferably is tribody or scDb-scFv.
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- 30 10. The multispecific antibody of any one of the preceding items, wherein said antibody does not comprise an immunoglobulin Fc region polypeptide, and, optionally, does not comprise CH1 and/or CL regions.
11. The multispecific antibody of any one of the preceding items, wherein said antibody is a tribody.
12. The multispecific antibody of item 11, wherein said first domain, said second domain and said third domain are independently selected from the group consisting of Fab and

scFv, preferably wherein said second domain is Fab and said first and third domains are scFvs.

13. The multispecific antibody of item 9, wherein said antibody is an scDb-scFv, preferably wherein said scFv moiety is C-terminally fused to the scDb, more preferably wherein said first domain and said second domain form an scDb and said third domain is an scFv.

14. The multispecific antibody of item 13, wherein said antibody is represented by the formula:

VLA - L1 - VHC - L2 - VLC - L3 - VHA - L4 - VLB - L5 - VHB or

VLB - L1 - VHA - L2 - VLA - L3 - VHB - L4 - VLC - L5 - VHC or

VLC - L1 - VHB - L2 - VLB - L3 - VHC - L4 - VLA - L5 - VHA or

VLA - L1 - VHB - L2 - VLB - L3 - VHA - L4 - VLC - L5 - VHC,

preferably VLB - L1 - VHA - L2 - VLA - L3 - VHB - L4 - VLC - L5 - VHC or VLA - L1 - VHB - L2 - VLB - L3 - VHA - L4 - VLC - L5 - VHC,

more preferably VLB - L1 - VHA - L2 - VLA - L3 - VHB - L4 - VLC - L5 - VHC,

wherein VLA and VHA are a light chain variable region and a heavy chain variable region of the first domain, respectively; and VLB and VHB are a light chain variable region and a heavy chain variable region of the second domain, respectively; and VLC and VHC are a light chain variable region and a heavy chain variable region of the third domain, respectively, and wherein L1, L2, L3, L4 and L5 are polypeptide linkers.

15. The multispecific antibody of item 14, wherein said L1 and L3 are as set forth in SEQ ID NO: 132.

16. The multispecific antibody of any one of items 14 to 15, wherein said L2, L4 and L5 are as set forth in SEQ ID NO: 23.

17. The multispecific antibody of any one of the preceding items, wherein said antibody has the following properties

(a) has the ability to neutralize IL-17A with a potency relative to that of secukinumab (relative potency), determined by measuring Gro- α secretion in an HT-29 assay, greater than 2, e.g., greater than 5, greater than 10, greater than 15, greater than 20, greater than 25, greater than 30, greater than 35, greater than 40, greater than 45, preferably greater than 50, and wherein said relative potency is the ratio of the IC₅₀

- value in ng/mL of secukinumab as measured in the HT-29 assay to the IC₅₀ value in ng/mL of said multispecific antibody as measured in the HT-29 assay; and
- (b) has the ability to neutralize TNF α with a potency relative to that of an scDb according to SEQ ID NO: 149 (A13) (relative potency), determined by measuring Gro- α secretion in an HT-29 assay, of at least 1, e.g., greater than 1, greater than 1.5, greater than 2, greater than 2.5, greater than 3, greater than 3.5, preferably greater than 4, more preferably greater than 4.5, and wherein said relative potency is the ratio of the IC₅₀ value in nM of said scDb according to SEQ ID NO: 149 as measured in the HT-29 assay to the IC₅₀ value in nM of said multispecific antibody as measured in the HT-29 assay; and
- (c) optionally, has a the ability to block interaction between IL-17A and IL-17RA with a potency relative to that of secukinumab (relative potency), determined in ELISA assay, greater than 2, e.g., greater than 3, greater than 4, greater than 5, greater than 6, greater than 7, greater than 8, greater than 9, preferably greater than 10, and wherein said relative potency is the ratio of the IC₅₀ value in ng/mL of secukinumab as measured by ELISA to the IC₅₀ value in ng/mL of said multispecific antibody as measured by ELISA; and
- (d) optionally, has the ability to neutralize TNF α with a potency relative to that of an scDb according to SEQ ID NO: 149 (A13) (relative potency), determined in L929 assay, of at least 0.4, e.g., at least 0.5, preferably at least 1, and wherein said relative potency is the ratio of the IC₅₀ value in nM of said scDb according to SEQ ID NO: 149 as measured in the L929 assay to the IC₅₀ value in nM of said multispecific antibody as measured in the L929 assay; and/or
- (e) binds to human IL-17A with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, less than 2 nM, than 1 nM, preferably less than 0.5 nM, as measured by surface plasmon resonance; and optionally, binds to cynomolgus IL-17A with a K_D of less than 5 nM, e.g., less than 4 nM, less than 3 nM, less than 2 nM, than 1 nM, preferably less than 0.5 nM as measured by surface plasmon resonance;
- (f) binds to human TNF α with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, less than 2 nM, less than 1 nM, preferably less than 0.5 nM, more preferably less than 0.25 nM, as measured by surface plasmon resonance; and

- (g) optionally, binds to human serum albumin with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, preferably less than 2 nM, as measured by surface plasmon resonance, and optionally, binds to cynomolgus serum albumin with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, preferably less than 2 nM as measured by surface plasmon resonance.
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18. The multispecific antibody of any one of the preceding items, wherein said antibody has the following properties
- (a) has a melting temperature (T_m), determined by differential scanning fluorimetry, of at least 55°C, preferably of at least 58°C, more preferably at least 60°C in phosphate-citrate buffer at pH 6.4, 150 mM NaCl;
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- (b) has a loss in monomer content, after five consecutive freeze-thaw cycles, of less than 5%, e.g., less than 4%, less than 3%, less than 2%, preferably 1% or less, when said multispecific antibody is at a starting concentration of 10 mg/ml in phosphate buffered saline (PBS), pH 7.4;
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- (c) has a loss in monomer content, after storage for at least two weeks, particularly for at least four weeks, at 4°C, of less than 10%, preferably less than 5%, when said multispecific antibody is at a starting concentration of 10 mg/ml in phosphate buffered saline (PBS), pH 7.4; and/or
- 20
- (d) has a loss in monomer content, after storage for at least two weeks, particularly for at least four weeks, at 37°C, of less than 20%, preferably less than 15%, when said multispecific antibody is at a starting concentration of 10 mg/ml in phosphate buffered saline (PBS), pH 7.4.
19. The multispecific antibody of any one of the preceding items wherein each domain comprises a heavy chain variable region (VH) and a light chain variable region (VL), wherein:
- 25
- (a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, and
- (b) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3.
- 30
20. The multispecific antibody of any one of the preceding items, wherein said first domain specifically binding IL-17A comprises a set of CDRs: HCDR1, HCDR2, HCDR3, LCDR1, LCDR2 and LCDR3, wherein the set of CDRs has 10 or fewer

amino acid substitutions from a set of CDRs in which (i) HCDR1' is as set forth in SEQ ID NO: 1; HCDR2' is as set forth in SEQ ID NO: 2; HCDR3' is as set forth in SEQ ID NO: 3; LCDR1' is as set forth in SEQ ID NO: 12; LCDR2' is as set forth in SEQ ID NO: 13; LCDR3' is as set forth in SEQ ID NO: 14, or (ii) HCDR1' is as set forth in SEQ ID NO: 39; HCDR2' is as set forth in SEQ ID NO: 40; HCDR3' is as set forth in SEQ ID NO: 41; LCDR1' is as set forth in SEQ ID NO: 50; LCDR2' is as set forth in SEQ ID NO: 51; LCDR3' is as set forth in SEQ ID NO: 52.

21. The multispecific antibody of item 20 wherein

- (i)
 - 10 (a) said HCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 1, 4, and 7;
 - (b) said HCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 2, 5, and 8;
 - (c) said HCDR3 is as set forth in the amino acid sequence selected from any one of
 - 15 SEQ ID Nos: 3, 6, and 9;
 - (d) said LCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 12, 15, and 18;
 - (e) said LCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 13, 16, and 19; and
 - 20 (f) said LCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 14, 17, and 20; or
- (ii)
 - (a) said HCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45;
 - 25 (b) said HCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 40, 43, and 46;
 - (c) said HCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 41, 44, and 47;
 - (d) said LCDR1 is as set forth in the amino acid sequence selected from any one of
 - 30 SEQ ID Nos: 50, 53, and 56;
 - (e) said LCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 51, 54, and 57; and

- (f) said LCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 52, 55, and 58.
22. The multispecific antibody of item 21 comprising (i) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively, or (ii) (i) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively.
23. The multispecific antibody of any one of the preceding items, wherein said first domain specifically binding IL-17A comprises a heavy chain variable region VHA and wherein said VHA is VH3 or VH4, preferably VH3.
24. The multispecific antibody of any one of the preceding items, wherein said first domain specifically binding IL-17A comprises a light chain variable region VLA and wherein said VLA comprises V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 FR1 to FR3, and a framework FR4, which is selected from a V λ FR4, particularly V λ 1 FR4, V λ 3 FR4, and V λ FR4, particularly V λ FR4 comprising the amino acid sequence having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in SEQ ID NO: 26 or 27, more preferably V λ FR4 as set forth in SEQ ID NO: 27.
25. The multispecific antibody of any one of items 23 to 24, wherein said VHA comprises an amino acid sequence that is (i) at least 90 percent identical to the amino acid sequence SEQ ID NO: 10; and/or said VLA comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 21, or (ii) at least 90 percent identical to the amino acid sequence SEQ ID NO: 48; and/or said VLA comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 59.
26. The multispecific antibody of item 25, wherein said VHA comprises an amino acid sequence (i) selected from the group consisting of SEQ ID NOs: 10 and 11; and/or said VLA comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 21 and 22, or (ii) selected from the group consisting of SEQ ID NOs: 48

- and 49; and/or said VLA comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 59 and 60.
27. The multispecific antibody of item 26, comprising (i) a VHA sequence of SEQ ID NO: 10 and/or a VLA sequence of SEQ ID NO: 21, or (ii) a VHA sequence of SEQ ID NO: 48 and/or a VLA sequence of SEQ ID NO: 59.
28. The multispecific antibody of any one of the preceding items, wherein said second domain specifically binding TNF α comprises a set of CDRs: HCDR1, HCDR2, HCDR3, LCDR1, LCDR2 and LCDR3, wherein the set of CDRs has 10 or fewer amino acid substitutions from a set of CDRs in which HCDR1' is as set forth in SEQ ID NO: 63; HCDR2' is as set forth in SEQ ID NO: 64; HCDR3' is as set forth in SEQ ID NO: 65; LCDR1' is as set forth in SEQ ID NO: 76; LCDR2' is as set forth in SEQ ID NO: 77; LCDR3' is as set forth in SEQ ID NO: 78.
29. The multispecific antibody of item 28, wherein
- (a) said HCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 63, 66, and 69;
- (b) said HCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 64, 67, and 70;
- (c) said HCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 65, 68, and 71;
- (d) said LCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 76, 79, and 82;
- (e) said LCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 77, 80, and 83; and
- (f) said LCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 78, 81, and 84.
30. The multispecific antibody of item 29 comprising HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 63, 64, and 65, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 76, 77, and 78, respectively.
31. The multispecific antibody of any one of the preceding items, wherein said second domain specifically binding TNF α comprises a heavy chain variable region VHB and wherein said VHB is VH3 or VH4, preferably VH3.
32. The multispecific antibody of any one of the preceding items, wherein said second domain specifically binding TNF α comprises a light chain variable region VLB and

- wherein said VLB comprises V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 FR1 to FR3, and a framework FR4, which is selected from a V κ FR4, particularly V κ 1 FR4, V κ 3 FR4, and V λ FR4, particularly V λ FR4 comprising the amino acid sequence having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in SEQ ID NO: 26 or 27, more preferably V λ FR4 as set forth in SEQ ID NO: 27.
33. The multispecific antibody of any one of items 31 to 32, wherein said VHB comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence selected from the group consisting of SEQ ID NOs: 72, 73, 74 and 75, preferably at least 90 percent identical to SEQ ID NO: 72; and/or said VLB comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence selected from the group consisting of SEQ ID NOs: 85, 86, 87, 88 and 89, preferably at least 90 percent identical to SEQ ID NO: 85.
34. The multispecific antibody of item 33, wherein said VHB comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 72, 73, 74 and 75; and/or said VLB comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 85, 86, 87, 88 and 89.
35. The multispecific antibody of item 34, comprising (i) a VHB sequence of SEQ ID NO: 72 and/or a VLB sequence of SEQ ID NO: 85; or (ii) a VHB sequence of SEQ ID NO: 75 and/or a VLB sequence of SEQ ID NO: 88; or (iii) a VHB sequence of SEQ ID NO: 75 and/or a VLB sequence of SEQ ID NO: 89.
36. The multispecific antibody of any one of items 6 to 35, wherein said third domain specifically binding HSA comprises a set of CDRs: HCDR1, HCDR2, HCDR3, LCDR1, LCDR2 and LCDR3, wherein the set of CDRs has 10 or fewer amino acid substitutions from a set of CDRs in which
- (a) HCDR1' is as set forth in SEQ ID NO: 90; HCDR2' is as set forth in SEQ ID NO: 91; HCDR3' is as set forth in SEQ ID NO: 92; LCDR1' is as set forth in SEQ ID NO: 100; LCDR2' is as set forth in SEQ ID NO: 101; LCDR3' is as set forth in SEQ ID NO: 102; or
- (b) HCDR1' is as set forth in SEQ ID NO: 111; HCDR2' is as set forth in SEQ ID NO: 112; HCDR3' is as set forth in SEQ ID NO: 113; LCDR1' is as set forth in SEQ ID

NO: 120; LCDR2' is as set forth in SEQ ID NO: 121; LCDR3' is as set forth in SEQ ID NO: 122.

37. The multispecific antibody of item 36, wherein

- 5 (a) said HCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 90, 93, and 96; said HCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 91, 94, and 97; said HCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 92, 95, and 98; said LCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 100, 103, and 106; said LCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 101, 104, and 107; and said LCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 102, 105, and 108; or
- 10 (b) said HCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 111, 114, and 117; said HCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 112, 115, and 118; said HCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 113, 116, and 119; said LCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 121, 124, and 127; said LCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 122, 125, and 128; and said LCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 123, 126, and 129.

38. The multispecific antibody of item 37 comprising

- 25 (a) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 90, 91, and 92, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 100, 101, and 102, respectively; or
- (b) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 111, 112, and 113, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 121, 122, and 123, respectively.

39. The multispecific antibody of any one of items 6 to 38, wherein said third domain specifically binding HSA comprises a heavy chain variable region VHC and wherein said VHC is VH3 or VH4, preferably VH3.

40. The multispecific antibody of any one of items 6 to 39, wherein said third domain specifically binding HSA comprises a light chain variable region VLC and wherein

- said VLC comprises V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 FR1 to FR3, and a framework FR4, which is selected from a V κ FR4, particularly V κ 1 FR4, V κ 3 FR4, and V λ FR4, particularly V λ FR4 comprising the amino acid sequence having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in SEQ ID NO: 26 or 27, more preferably V λ FR4 as set forth in SEQ ID NO: 27.
- 5
41. The multispecific antibody of any one of items 39 to 40, wherein said VHC comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 99; and/or said VLC comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 109.
- 10
42. The multispecific antibody of item 41, comprising a VHC sequence of SEQ ID NO: 99 and/or a VLC sequence of SEQ ID NO: 109.
- 15
43. The multispecific antibody of any one of items 39 to 40, wherein said VHC comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 110; and/or said VLC comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 120.
44. The multispecific antibody of item 43, comprising a VHC sequence of SEQ ID NO: 110 and/or a VLC sequence of SEQ ID NO: 120.
- 20
45. The multispecific antibody of any one of the preceding items, wherein the antibody is humanized.
46. The multispecific antibody of any of the preceding items, wherein the antibody comprises an amino acid sequence having at least 80% identity, preferably at least 90% identity, to the sequence selected from any of SEQ ID NOs: 136, 137, 138, 139, 140, 141, 142, 143, 144, 145, 146, 147, and 148, preferably 143, wherein CDRs have the sequences according to items 22, 30 and 38(a).
- 25
47. The multispecific antibody of any of the preceding items, wherein the antibody comprises an amino acid sequence selected from any of SEQ ID NOs: 136, 137, 138, 139, 140, 141, 142, 143, 144, 145, 146, 147, and 148, preferably 143.
- 30
48. A pharmaceutical composition comprising the multispecific antibody of any one of the preceding items and a pharmaceutically acceptable carrier.

49. The multispecific antibody of any one of items 1 to 47, or the pharmaceutical composition of item 48 for use as a medicament.
50. The multispecific antibody of any one of items 1 to 47, or the pharmaceutical composition of item 48 for use in the treatment of a disorder mediated by IL-17A and/or TNF α or a disorder that can be treated by inhibiting Gro- α secretion.
51. The multispecific antibody of any one of items 1 to 47, or the pharmaceutical composition of item 48 for use in the treatment of an inflammatory condition or an autoimmune disease.
52. The multispecific antibody of any one of items 1 to 47, or the pharmaceutical composition of item 48 for use in the treatment of a cancer, arthritis, rheumatoid arthritis, osteoarthritis, reactive arthritis, psoriasis, chronic obstructive pulmonary disease, systemic lupus erythematosus (SLE), lupus nephritis, an autoimmune inflammatory bowel disease, asthma, multiple sclerosis, or cystic fibrosis, bone loss, airways hypersensitivity, a demyelinating disorder, dermal hypersensitivity, acute transplant rejection, allograft rejection, graft-versus host disease, systemic sclerosis, an urological inflammatory disorder, a cardiovascular disease, vasculitis, a periodic fever, a glucose metabolism disorder, a pulmonary disease, peridontitis, hepatic stromal keratitis, an allergy, inflammatory pain, a spondyloarthropathy, septicaemia, septic or endotoxic shock, meningitis, surgical trauma, an autoimmune haematological disorder, Alzheimer's disease, sarcoidosis, cirrhosis, hepatitis, glomerulonephritis or dislipidemia.
53. Use of the multispecific antibody of any one of items 1 to 47, or the pharmaceutical composition of item 48 in the manufacture of a medicament for use in the treatment of a disorder mediated by IL-17A and/or TNF α or a disorder that can be treated by inhibiting Gro- α secretion.
54. Use of the multispecific antibody of any one of items 1 to 47, or the pharmaceutical composition of item 48 in the manufacture of a medicament for use in the treatment of an inflammatory condition or an autoimmune disease.
55. Use of the multispecific antibody of any one of items 1 to 47, or the pharmaceutical composition of item 48 in the manufacture of a medicament for use in the treatment of a cancer, arthritis, rheumatoid arthritis, osteoarthritis, reactive arthritis, psoriasis, chronic obstructive pulmonary disease, systemic lupus erythematosus (SLE), lupus nephritis, an autoimmune inflammatory bowel disease, asthma, multiple sclerosis,

- cystic fibrosis, bone loss, airways hypersensitivity, a demyelinating disorder, dermal hypersensitivity, acute transplant rejection, allograft rejection, graft-versus host disease, systemic sclerosis, an urological inflammatory disorder, a cardiovascular disease, vasculitis, a periodic fever, a glucose metabolism disorder, a pulmonary disease, peridontitis, hepatic stromal keratitis, an allergy, inflammatory pain, a spondyloarthropathy, septicaemia, septic or endotoxic shock, meningitis, surgical trauma, an autoimmune haematological disorder, Alzheimer's disease, sarcoidosis, cirrhosis, hepatitis, glomerulonephritis or dislipidemia.
56. A method of treating a disorder mediated by IL-17A and/or TNF α , said method comprising administering an effective amount of the multispecific antibody of any one of items 1 to 47, or the pharmaceutical composition of item 48, such that the condition is alleviated.
57. The method according to item 56, wherein the disorder mediated by IL-17A and/or TNF α is inflammatory condition or an autoimmune disease.
58. The method according to item 56, wherein the disorder mediated by IL-17A and/or TNF α is a cancer, arthritis, rheumatoid arthritis, osteoarthritis, reactive arthritis, psoriasis, chronic obstructive pulmonary disease, systemic lupus erythematosus (SLE), lupus nephritis, an autoimmune inflammatory bowel disease, asthma, multiple sclerosis, cystic fibrosis, bone loss, airways hypersensitivity, a demyelinating disorder, dermal hypersensitivity, acute transplant rejection, allograft rejection, graft-versus host disease, systemic sclerosis, an urological inflammatory disorder, a cardiovascular disease, vasculitis, a periodic fever, a glucose metabolism disorder, a pulmonary disease, peridontitis, hepatic stromal keratitis, an allergy, inflammatory pain, a spondyloarthropathy, septicaemia, septic or endotoxic shock, meningitis, surgical trauma, an autoimmune haematological disorder, Alzheimer's disease, sarcoidosis, cirrhosis, hepatitis, glomerulonephritis or dislipidemia.
59. A nucleic acid encoding the multispecific antibody of any one of items 1 to 47 or a fragment thereof.
60. A vector comprising the nucleic acid of item 59.
61. A host cell comprising the nucleic acid of item 59 or the vector of item 60.
62. A method of producing the multispecific antibody of any one of items 1 to 47, the method comprising the step of culturing a host cell comprising a nucleic acid or a

vector encoding the multispecific antibody of any one of items 1 to 47 or a fragment thereof.

63. A kit comprising the multispecific antibody of any one of items 1 to 47, or the pharmaceutical composition of item 48.

5 It is another object of the present invention to provide an anti-IL-17A antibody with improved affinity, efficacy and improved biophysical properties, e.g. improved solubility, developability and stability.

Anti-IL-17A antibodies of the present invention have improved properties beneficial for use in therapies, such as higher affinity, improved efficacy, selectivity, improved biophysical
10 properties, such as solubility, developability, and stability.

Therefore, in one aspect, the disclosure provides an isolated antibody having a binding specificity for human IL-17A, in particular which comprises a set of CDRs: HCDR1, HCDR2, HCDR3, LCDR1, LCDR2 and LCDR3, wherein the set of CDRs has 10 or fewer, preferably 0, amino acid substitutions from a set of CDRs in which HCDR1' is amino acid
15 sequence selected from any one of SEQ ID Nos: 1, 4, and 7; HCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 2, 5, and 8; HCDR3' is amino acid sequence selected from any one of SEQ ID Nos: 3, 6, and 9; LCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 12, 15, and 18; LCDR2' is amino acid sequence selected from any one of
20 SEQ ID Nos: 13, 16, and 19; LCDR3' having the amino acid sequence selected from any one of SEQ ID Nos: 14, 17, and 20. In one aspect, the present disclosure relates to a multispecific molecule comprising the isolated antibody of the disclosure.

In another aspect, the disclosure provides an isolated antibody having a binding specificity for human IL-17A, in particular which comprises a set of CDRs: HCDR1, HCDR2, HCDR3, LCDR1, LCDR2 and LCDR3, wherein the set of CDRs has 10 or fewer,
25 preferably 0, amino acid substitutions from a set of CDRs in which HCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45; HCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 40, 43, and 46; HCDR3' is amino acid sequence selected from any one of SEQ ID Nos: 41, 44, and 47; LCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 50, 53, and 56; LCDR2' is amino acid
30 sequence selected from any one of SEQ ID Nos: 51, 54, and 57; LCDR3' having the amino

acid sequence selected from any one of SEQ ID Nos: 52, 55, and 58. In one aspect, the present disclosure relates to a multispecific molecule comprising the isolated antibody of the disclosure.

5 In one aspect, the present disclosure relates to a pharmaceutical composition comprising the isolated antibody of the disclosure, or the multispecific molecule comprising the isolated antibody of the disclosure, and a pharmaceutically acceptable carrier.

In another aspect, the present disclosure relates to the antibody of the disclosure or the multispecific molecule comprising said isolated antibody, or the pharmaceutical composition of the disclosure for use as a medicament.

10 In one aspect, the present disclosure relates to the antibody, or the multispecific molecule comprising said isolated antibody, or the pharmaceutical composition of the disclosure for use in the treatment of a disorder mediated by IL-17A or a disorder that can be treated by inhibiting GRO- α secretion.

15 In one aspect, the present disclosure relates to use of the antibody of the disclosure or the multispecific molecule comprising said isolated antibody, or the pharmaceutical composition of the disclosure in the manufacture of a medicament for use in the treatment of a disorder mediated by IL-17A or a disorder that can be treated by inhibiting GRO- α secretion.

20 In another aspect, the present disclosure relates to a method of treating a disorder mediated by IL-17A, said method comprising administering an effective amount of the antibody of the disclosure, or the multispecific molecule of the disclosure, or the pharmaceutical composition of the disclosure to a subject in need thereof. In yet another aspect, the present disclosure relates to a nucleic acid encoding the antibody of the disclosure. In a further aspect, the present disclosure relates to a vector comprising said nucleic acid. In a further aspect, the present disclosure relates to a host cell comprising said nucleic acid or said
25 vector.

In another aspect, the present disclosure relates to a method of producing the antibody of the disclosure, the method comprising the step of culturing a host cell comprising a nucleic acid or a vector encoding the antibody of the disclosure.

The aspects, advantageous features and preferred embodiments of the present disclosure summarized in the following items, respectively alone or in combination, further contribute to solving the object of the invention:

1. An antibody having a binding specificity for human IL-17A, comprising a set of
5 CDRs: HCDR1, HCDR2, HCDR3, LCDR1, LCDR2 and LCDR3, wherein the set of CDRs has 10 or fewer amino acid substitutions from a set of CDRs in which
(i) HCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 1, 4, and 7;
HCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 2, 5, and 8;
HCDR3' is amino acid sequence selected from any one of SEQ ID Nos: 3, 6, and 9;
10 LCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 12, 15, and 18;
LCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 13, 16, and 19;
LCDR3' having the amino acid sequence selected from any one of SEQ ID Nos: 14,
15 17, and 20; or
(ii) HCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45;
HCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 40, 43, and 46;
20 HCDR3' is amino acid sequence selected from any one of SEQ ID Nos: 41, 44, and 47;
LCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 50, 53, and 56;
LCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 51, 54, and
25 57;
LCDR3' having the amino acid sequence selected from any one of SEQ ID Nos: 52, 55, and 58.
2. The antibody of item 1, comprising a set of CDRs: HCDR1, HCDR2, HCDR3,
LCDR1, LCDR2 and LCDR3, wherein the set of CDRs has 10 or fewer amino acid
30 substitutions from a set of CDRs in which
(i) HCDR1' is as set forth in SEQ ID Nos: 1;
HCDR2' is as set forth in SEQ ID Nos: 2;

- HCDR3' is as set forth in SEQ ID Nos: 3;
LCDR1' is as set forth in SEQ ID Nos: 12;
LCDR2' is as set forth in SEQ ID Nos: 13;
LCDR3' is as set forth in SEQ ID Nos: 14; or
5 (ii) HCDR1' is as set forth in SEQ ID Nos: 39;
HCDR2' is as set forth in SEQ ID Nos: 40;
HCDR3' is as set forth in SEQ ID Nos: 41;
LCDR1' is as set forth in SEQ ID Nos: 50;
LCDR2' is as set forth in SEQ ID Nos: 51;
10 LCDR3' is as set forth in SEQ ID Nos: 52.
3. The antibody of item 1 or item 2, which comprises a heavy chain variable region (VH) and a light chain variable region (VL), wherein:
- (c) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, and
15 (d) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3.
4. The antibody of item 3, wherein
- (i)
- (g) said HCDR1 is as set forth in the amino acid sequence selected from any one of
20 SEQ ID Nos: 1, 4, and 7;
(h) said HCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 2, 5, and 8;
(i) said HCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 3, 6, and 9;
25 (j) said LCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 12, 15, and 18;
(k) said LCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 13, 16, and 19; and
(l) said LCDR3 is as set forth in the amino acid sequence selected from any one of
30 SEQ ID Nos: 14, 17, and 20; or
- (ii)

- (a) said HCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45;
- (b) said HCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 40, 43, and 46;
- 5 (c) said HCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 41, 44, and 47;
- (d) said LCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 50, 53, and 56;
- (e) said LCDR2 is as set forth in the amino acid sequence selected from any one of
- 10 SEQ ID Nos: 51, 54, and 57; and
- (f) said LCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 52, 55, and 58.
5. The antibody of item 4, wherein the antibody comprises (i) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and LCDR1, LCDR2,
- 15 and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (ii) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively.
6. The antibody of any one of items 3 to 5, wherein said VH is VH3 or VH4, preferably
- 20 VH3.
7. The antibody of any one of items 3 to 6, wherein said VL comprises V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 FR1 to FR3, and a framework FR4, which is selected from a V κ FR4, particularly V κ 1 FR4, V κ 3 FR4, and V λ FR4, particularly V λ FR4 comprising the amino acid sequence having at
- 25 least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in SEQ ID NO: 26 or 27, more preferably V λ FR4 as set forth in SEQ ID NO: 27.
8. The antibody of any one of items 3 to 7, wherein (i) said VH comprises an amino acid
- 30 sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 10; and/or said VL comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 21; or (i) said VH comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence

SEQ ID NO: 48; and/or said VL comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 59.

9. The antibody of item 8, wherein (i) said VH comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 10 and 11; and/or said VL comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 21 and 22; or (ii) said VH comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 48 and 49; and/or said VL comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 59 and 60.
10. The antibody of item 9, comprising (i) a VH sequence of SEQ ID NO: 10 and/or a VL sequence of SEQ ID NO: 21; or (ii) a VH sequence of SEQ ID NO: 48 and/or a VL sequence of SEQ ID NO: 59.
11. The antibody of item 9, comprising (i) a VH sequence of SEQ ID NO: 11 and/or a VL sequence of SEQ ID NO: 49; or (ii) a VH sequence of SEQ ID NO: 11 and/or a VL sequence of SEQ ID NO: 60.
12. The antibody of any one of the preceding items, wherein the antibody has a binding specificity for cynomolgus monkey IL-17A.
13. The antibody of any one of the preceding items, wherein said antibody selectively binds to human IL-17A over human IL-17B, IL-17C, IL-17D, IL-17E and IL-17F as measured by ELISA.
14. The antibody of any one of the preceding items, wherein binding to IL-17A
- (a) inhibits or blocks binding between IL-17A and its receptor (IL-17RA), and
 - (b) reduces or neutralizes IL-17A activity.
15. The antibody of item 14, wherein said antibody is capable of inhibiting GRO- α secretion when assessed *in vitro* in HT-29 assay.
16. The antibody of any of the preceding items, wherein said antibody:
- (h) has the ability to block interaction between IL-17A and IL-17RA with a potency relative to that of secukinumab (relative potency), determined in ELISA assay, greater than 5, preferably greater than 10, more preferably greater than 15, even more preferably greater than 20, and wherein said relative potency is the ratio of the IC₅₀ value in ng/mL of secukinumab as measured by ELISA to the IC₅₀ value in ng/mL of the antibody of the invention in the scFv format as measured by ELISA; and/or

- (i) has the ability to neutralize IL-17A with a potency relative to that of secukinumab (relative potency), determined by measuring GRO- α secretion in an HT-29 assay, greater than 50, preferably greater than 100, more preferably greater than 150, and wherein said relative potency is the ratio of the IC₅₀ value in ng/mL of
5 secukinumab as measured in the HT-29 assay to the IC₅₀ value in ng/mL of the antibody of the invention in the scFv format as measured in the HT-29 assay; and/or
- (j) capable of inhibiting the activity of 1 ng human IL-17A at a concentration of 1 ng/mL or less, preferably 0.5 ng/mL or less, more preferably 0.2 ng/mL or less, by
10 50%, said inhibitory activity is determined by measuring GRO- α secretion induced by human IL-17A in HT-29 assay in the presence of 50 pg/ml TNF α .
17. The antibody of any of the preceding items, wherein said antibody:
- (a) binds to human IL-17A with a dissociation constant (K_D) of less than 5 nM, particularly less than 1 nM, less than 0.5 nM, less than 0.2 nM, more
15 particularly less than 100 pM, more particularly less than 50 pM, as measured by surface plasmon resonance, particularly as measured by surface plasmon resonance in a direct setup; and
- (b) optionally, binds to Cynomolgus IL-17A with a K_D of less than 10 nM, particularly less than 7 nM, less than 5 nM, less than 2 nM, less than 1 nM,
20 more particularly less than 0.5 nM as measured by surface plasmon resonance, particularly as measured by surface plasmon resonance in a capture setup.
18. The antibody of item 17, wherein said antibody binds to human IL-17A with a dissociation constant (K_D) of less than 0.5 nM, less than 0.2 nM, less than 100 pM, particularly less than 50 pM as measured by surface plasmon resonance, in particular
25 as measured by surface plasmon resonance in a direct set-up.
19. The antibody of any of the preceding items, wherein said antibody:
- (e) when in scFv format, has a melting temperature (T_m), determined by differential scanning fluorimetry, of at least 60°C, particularly of at least 62°C, of at least 65°C, more particularly of at least 70°C, in particular
30 wherein said antibody is in phosphate-citrate buffer at pH 6.4, 150 mM NaCl;
- (f) when in scFv format, has a loss in monomer content, after five consecutive freeze-thaw cycles, of less than 5%, particularly less than 3%, more

- particularly less than 1%, when the antibody of the invention is at a starting concentration of 10 mg/ml, in particular wherein said antibody is in phosphate buffered saline (PBS), pH 7.4;
- (g) when in scFv format, has a loss in monomer content, after storage for at least two weeks, particularly for at least four weeks, at 4°C, of 5% or less, particularly less than 4%, less than 3%, less than 2%, more particularly less than 1%, when the antibody of the invention is at a starting concentration of 10 mg/ml, in particular wherein said antibody is in phosphate buffered saline (PBS), pH 7.4; and/or
- (h) has a loss in monomer content, after storage for at least two weeks, particularly for at least four weeks, at 37°C, of less than 5%, when the antibody of the invention is at a starting concentration of 10 mg/ml.
20. The antibody of any of the preceding items, wherein the antibody is selected from the group consisting of: a monoclonal antibody, a chimeric antibody, a Fab, an Fv, an scFv, dsFv, an scAb, STAB, a single domain antibody (sdAb or dAb), a single domain heavy chain antibody, and a single domain light chain antibody, a VHH, a VNAR, single domain antibodies based on the VNAR structure from shark, and binding domains based on alternative scaffolds including but limited to ankyrin-based domains, fynomers, avimers, anticalins, fibronectins, and binding sites being built into constant regions of antibodies (e.g. F-star's Modular Antibody TechnologyTM), preferably scFv.
21. The antibody of item 20, wherein said scFv has the amino acid sequence selected from (i) the group consisting of SEQ ID NO:24 and SEQ ID NO: 25, preferably wherein said scFv has the amino acid sequence of SEQ ID NO: 24; or (ii) the group consisting of SEQ ID NO:61 and SEQ ID NO: 62, preferably wherein said scFv has the amino acid sequence of SEQ ID NO: 61.
22. The isolated antibody of item 20, wherein the antibody is an IgG selected from the group consisting of an IgG1, an IgG2, an IgG3 and an IgG4, preferably wherein the antibody is an IgG1 or IgG4.
23. The isolated antibody of any of the preceding items, wherein the antibody is humanized.
24. The antibody of any one of items 1 to 23 which is a multispecific molecule.

25. The antibody of item 24, wherein said antibody is in a format selected from the group consisting of a single-chain diabody (scDb), a tandem scDb (Tandab), a linear dimeric scDb (LD-scDb), a circular dimeric scDb (CD-scDb), a bispecific T-cell engager (BiTE; tandem di-scFv), a tandem tri-scFv, a tribody (Fab-(scFv)₂) or bibody (Fab-(scFv)₁), Fab, Fab-Fv₂, Morrison (IgG CH₃-scFv fusion (Morrison L) or IgG CL-scFv fusion (Morrison H)), triabody, scDb-scFv, bispecific Fab₂, di-miniantibody, tetraabody, scFv-Fc-scFv fusion, scFv-HSA-scFv fusion, di-diabody, DVD-Ig, COVD, IgG-scFab, scFab-dsscFv, Fv₂-Fc, IgG-scFv fusions, such as bsAb (scFv linked to C-terminus of light chain), Bs1Ab (scFv linked to N-terminus of light chain), Bs2Ab (scFv linked to N-terminus of heavy chain), Bs3Ab (scFv linked to C-terminus of heavy chain), Ts1Ab (scFv linked to N-terminus of both heavy chain and light chain), Ts2Ab (dsscFv linked to C-terminus of heavy chain), bispecific antibodies based on heterodimeric Fc domains, such as Knob-into-Hole antibodies (KiHs); an Fv, scFv, scDb, tandem-di-scFv, tandem tri-scFv, Fab-(scFv)₂, Fab-(scFv)₁, Fab, Fab-Fv₂, COVD fused to the N- and/or the C-terminus of either chain of a heterodimeric Fc domain or any other heterodimerization domain, a MATCH and DuoBodies.
26. A pharmaceutical composition comprising the antibody of any one of items 1 to 25, and a pharmaceutically acceptable carrier.
27. The antibody of any one of items 1 to 25, or the pharmaceutical composition of item 26 for use as a medicament.
28. The antibody of any one of items 1 to 25, or the pharmaceutical composition of item 26 for use in the treatment of a disorder mediated by IL-17A or a disorder that can be treated by inhibiting GRO- α secretion.
29. The antibody of any one of items 1 to 25, or the pharmaceutical composition of item 26 for use in the treatment of an inflammatory condition or an autoimmune disease.
30. The antibody of any one of items 1 to 25, or the pharmaceutical composition of item 26 for use in the treatment of a cancer, arthritis, rheumatoid arthritis, osteoarthritis, reactive arthritis, psoriasis, chronic obstructive pulmonary disease, systemic lupus erythematosus (SLE), lupus nephritis, an autoimmune inflammatory bowel disease, asthma, multiple sclerosis, or cystic fibrosis, bone loss, airways hypersensitivity, a demyelinating disorder, dermal hypersensitivity, acute transplant rejection, allograft rejection, graft-versus host disease, systemic sclerosis, an urological inflammatory disorder, a cardiovascular disease, vasculitis, a periodic fever, a glucose metabolism

disorder, a pulmonary disease, peridontitis, hepatic stromal keratitis, an allergy, inflammatory pain, a spondyloarthropathy, septicaemia, septic or endotoxic shock, meningitis, surgical trauma, an autoimmune haematological disorder, Alzheimer's disease, sarcoidosis, cirrhosis, hepatitis, glomerulonephritis or dislipidemia.

- 5 31. Use of the antibody of any one of items 1 to 25, or the pharmaceutical composition of item 26 in the manufacture of a medicament for use in the treatment of a disorder mediated by IL-17A or a disorder that can be treated by inhibiting GRO- α secretion.
32. Use of the antibody of any one of items 1 to 25, or the pharmaceutical composition of item 26 in the manufacture of a medicament for use in the treatment of an
10 inflammatory condition or an autoimmune disease.
33. Use of the antibody of any one of items 1 to 25, or the pharmaceutical composition of item 26 in the manufacture of a medicament for use in the treatment of a cancer, arthritis, rheumatoid arthritis, osteoarthritis, reactive arthritis, psoriasis, chronic obstructive pulmonary disease, systemic lupus erythematosus (SLE), lupus nephritis,
15 an autoimmune inflammatory bowel disease, asthma, multiple sclerosis, cystic fibrosis, bone loss, airways hypersensitivity, a demyelinating disorder, dermal hypersensitivity, acute transplant rejection, allograft rejection, graft-versus host disease, systemic sclerosis, an urological inflammatory disorder, a cardiovascular disease, vasculitis, a periodic fever, a glucose metabolism disorder, a pulmonary
20 disease, peridontitis, hepatic stromal keratitis, an allergy, inflammatory pain, a spondyloarthropathy, septicaemia, septic or endotoxic shock, meningitis, surgical trauma, an autoimmune haematological disorder, Alzheimer's disease, sarcoidosis, cirrhosis, hepatitis, glomerulonephritis or dislipidemia.
34. A method of treating a disorder mediated by IL-17A, said method comprising
25 administering an effective amount of the antibody of any one of items 1 to 25, or the pharmaceutical composition of item 26, such that the condition is alleviated.
35. The method according to item 34, wherein the disorder mediated by IL-17A is inflammatory condition or an autoimmune disease.
36. The method according to item 34, wherein the disorder mediated by IL-17A is a
30 cancer, arthritis, rheumatoid arthritis, osteoarthritis, reactive arthritis, psoriasis, chronic obstructive pulmonary disease, systemic lupus erythematosus (SLE), lupus nephritis, an autoimmune inflammatory bowel disease, asthma, multiple sclerosis, cystic fibrosis, bone loss, airways hypersensitivity, a demyelinating disorder, dermal

- hypersensitivity, acute transplant rejection, allograft rejection, graft-versus host disease, systemic sclerosis, an urological inflammatory disorder, a cardiovascular disease, vasculitis, a periodic fever, a glucose metabolism disorder, a pulmonary disease, peridontitis, hepatic stromal keratitis, an allergy, inflammatory pain, a spondyloarthropathy, septicaemia, septic or endotoxic shock, meningitis, surgical trauma, an autoimmune haematological disorder, Alzheimer's disease, sarcoidosis, cirrhosis, hepatitis, glomerulonephritis or dislipidemia.
- 5 37. A nucleic acid encoding the antibody of items 1-25.
38. A vector comprising the nucleic acid of item 37.
- 10 39. A host cell comprising the nucleic acid of item 37 or the vector of item 38.
40. A method of producing the antibody of items 1- 25, the method comprising the step of culturing a host cell comprising a nucleic acid or a vector encoding the antibody of items 1-25.
41. A kit comprising the antibody of any one of items 1 to 25, or the pharmaceutical composition of item 26.
- 15

The disclosure contemplates all combinations of any one or more of the foregoing aspects and/or embodiments, as well as combinations with any one or more of the embodiments set forth in the detailed description and examples.

Other features, objects, and advantages of the compositions and methods herein will be apparent from the description and drawings, and from the claims.

20

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 shows that labeled IL-17A is suitable for use in the sorting process. Biological activity of labeled IL-17A in the HT-29 assay. 3-fold serial dilutions of labeled and un-labeled IL-17A were tested in parallel for their potential to induce GRO- α secretion in HT-29 cells. EC₅₀ values for labeled (IL-17A-RPE) and unlabeled (IL-17A) IL-17A are 82.8 ng/ml and 55 ng/ml respectively.

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FIG. 2 shows the potencies of 27-07-G02 rabbit IgG (A), and 27-31-C04 IgG (B), to neutralize IL-17A in the HT-29 assay.

FIG. 3 shows the potencies of 27-07-G02 rabbit IgG (A), and 27-31-C04 IgG (B), to inhibit the interaction between IL-17A and IL-17RA.

FIG. 4 shows the potencies of the humanized scFvs A1 and A2 (A), and PRO571 and PRO592 (B), to neutralize IL-17A in the HT-29 assay.

5 **FIG. 5** shows the potencies of anti-IL-17A scFv A1 and A2 (A), and PRO571 and PRO592 (B), to inhibit the interaction between human IL-17A and IL-17RA (ELISA).

FIG. 6 shows the target specificity of the scFv A1 (A), and scFvs PRO571 and PRO592 (B), is shown. The potential to inhibit the interaction of biotinylated IL-17A with the scFvs by IL-17B to IL-17F was analyzed by competition ELISA. Dose-dependent effects of IL-17A and
10 IL-17B to IL-17F are shown.

FIG. 7 shows the thermal unfolding curves from DSF measurements of the scFvs A1 and A2 (A), and the scFv PRO571 and PRO592 (B). The resulting T_m values have been determined by fitting the data to a Boltzmann equation to obtain the midpoint of transition.

FIG. 8 shows the storage stability study the scFv A1 (A), and scFvs PRO571 and PRO592
15 (B), performed at a concentration of >10 mg/mL for 4 weeks at three temperatures (37°C, 4°C and -80°C). Monomeric content over time at different storage temperatures is shown on the left; protein concentration at different storage temperatures (4°C (left) and 37°C (right)) over the course of time is shown on the right. Monomeric content was determined by integration of SE-HPLC peak areas and protein concentrations were calculated by UV_{280} measurement.

20 **FIG. 9** shows the monitoring of monomeric content of the scFv A1 (A), and scFvs PRO571 and PRO592 (B), over 5 repeated freezing and thawing cycles.

FIG. 10 shows trispecific formats. A, Three domain permutation variants of Fab-(scFv)₂ molecules were designed. ScFv fusions at positions CL and CH1 of the tribody format are considered as equivalent, resulting in three variants of this format. B, three scDb-scFv domain
25 permutation variants were designed. Domain specificities are indicated at the bottom. Gly-Ser linkers joining variable domains and inter-domain disulfide bonds are indicated by a grey bracket.

FIG. 11 shows the lead manufacture generic process. A, overlay of SE-HPLC traces of final A3-A5 (left graph) and A6-A8 (right graph) samples. Peaks with a retention time of 7-8 minutes correspond to the apparent molecular weight of the monomer of the respective molecules; peaks with a retention time >10 minutes are buffer and salt related artefacts. B, SDS-PAGE analysis of A3-A8 under non-reducing (left) and reducing (right) buffer conditions. A molecular weight reference was loaded in middle lane. Bands under non-reducing conditions correspond to the expected molecular weight of ~100 kDa for A3-A5 and ~75 kDa for A6-A8, respectively. As expected, for the heterodimeric Fab-(scFv)₂ antibody format (A3-A5) bands shift to ~50 kDa under reducing conditions, while for A6-A8 bands at a molecular weight of ~75 kDa can be observed also under reducing conditions.

FIG. 12 shows the potency to neutralize TNF α in L929 assay. Absorbances measured using the cell counting kit-8 are presented in function of the trispecific molecules A3-A8 concentrations in nM. A13 (parental bispecific, HSA binder and TNF α blocker) was used as reference.

FIG. 13 shows a comparison between potency to neutralize human and cynomolgus TNF α . Absorbances measured using the cell counting kit-8 in presence of human or cynomolgus TNF- α are presented in function to the A5 and A7 concentrations.

FIG. 14 shows the concomitant blockade of TNF- α and IL-17A in HT-29 assay in presence of HSA. In vitro concomitant blockade of TNF α and IL-17A by six trispecific molecules A3-A8 was analyzed using the HT-29 cell-based assay in presence of 1 mg/ml of HSA. Secukinumab (IL-17A blocker) and A13 (parental bispecific, HSA binder and TNF α blocker) were used as references. GRO- α secretion data obtained are presented in function of molecule concentrations in nM (A, C and E) and in ng/ml (B, D and F). “No TNF α ” shows the GRO- α secretion upon addition of only IL-17A and corresponds to the maximum effect by TNF α blockade. “No IL17a” shows the GRO- α secretion upon addition of only TNF α and corresponds to the maximum effect by IL-17A blockade. “No IL17a, no TNF α ” shows the background GRO- α secretion when no IL-17A and TNF α is added and corresponds to the maximum effect by concomitant TNF α and IL-17A blockade.

FIG. 15 shows the neutralization of IL-17A binding to IL-17RA in competitive ELISA. Absorbance measured in the competitive ELISA assessing IL-17A binding to IL-17RA are

presented in function of increasing concentrations of the six trispecific molecules (A3-A8). Secukinumab (IL-17A blocker) was used as references.

FIG. 16 shows the simultaneous binding to human TNF α , human IL-17A and HSA by SPR. The six possible sequences of injections of the different analytes (human TNF α , human IL-17A and HSA) were performed on a MASS-1 SPR device and the obtained sensorgrams are shown. The trispecific molecules were immobilized on the sensor chip (A3 on channel 1B, Ch1B, A4 on channel 2B, Ch2B, A5 on channel 3B, Ch3B, A6 on channel 4B, Ch4B, A7 on channel 5B, Ch5B, A8 on channel 6B, Ch6B) and antigens were injected sequentially.

FIG. 17 shows a storage stability study performed at temperatures of 37°C, 4°C and -80°C at a protein concentration of 10 mg/mL for 4 weeks. Course of % monomeric content and % monomeric loss over time was recorded at d0, d2, d7, d14, d21 and d28.

FIG. 18 shows overlays of SE-HPLC traces of d0 (black, shaded) and d28 (grey) stability samples of A5 (left), A7 (middle) and A8 (right).

FIG. 19 shows the pharmacokinetic profile of A7 in cynomolgus monkey after intravenous (n=3) and subcutaneous (n=3) administration and ADA analysis.

FIG. 20 shows the schematic structure of Morrison L constructs A14 and A15.

FIG. 21 shows the average Size of soluble complexes determined by Dynamic Light Scattering for scDb-scFv A7.

FIG. 22 shows the average Size of soluble complexes determined by Dynamic Light Scattering for A14.

FIG. 23 shows the average Size of soluble complexes determined by Dynamic Light Scattering for A15.

FIG. 24 shows the protein concentration recovery of soluble complexes for scDb-scFv A7.

FIG. 25 shows the protein concentration recovery of soluble complexes for A14.

FIG. 26 shows the protein concentration recovery of soluble complexes for A15.

DETAILED DESCRIPTION OF THE INVENTION

The present disclosure is based on the discovery of multispecific antibody molecules that specifically bind to IL-17A and TNF α and have improved affinity, efficacy, and selectivity. In addition, the multispecific antibody of the disclosure has improved safety profile, as the
5 inventors have demonstrated that said antibody does not form immune complexes with TNF α , and thus has potentially low immunogenicity. Due to the bivalent binding of other multispecifics (e.g. Covagen, Abbvie, etc.) there is a high probability of immune complex formation that may result in immunogenicity or other adverse effects. In contrast, monovalent
10 bi- and trispecific constructs of the present disclosure have a reduced potential to form such complexes, and therefore a lower probability to lead to anti-drug antibodies and immune-related adverse effects. Moreover, the multispecific antibody of the present disclosure has improved biophysical properties, e.g., developability and producibility in high amount with relatively low impurities, and superior stability.

The present disclosure furthermore provides antibodies that specifically bind to human
15 IL-17A protein, and pharmaceutical compositions, production methods, and methods of use of such antibodies and pharmaceutical compositions.

Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by those of ordinary skill in the art to which this invention pertains.

20 The terms “comprising” and “including” are used herein in their open-ended and non-limiting sense unless otherwise noted. With respect to such latter embodiments, the term “comprising” thus includes the narrower term “consisting of”.

The terms “a” and “an” and “the” and similar references in the context of describing the invention (especially in the context of the following claims) are to be construed to cover both
25 the singular and the plural, unless otherwise indicated herein or clearly contradicted by context. For example, the term “a cell” includes a plurality of cells, including mixtures thereof. Where the plural form is used for compounds, salts, and the like, this is taken to mean also a single compound, salt, or the like.

Multispecific antibodies of the present disclosure

In one aspect, the disclosure provides an isolated multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α .

The term “TNF α ” or “TNF- α ” or “tumor necrosis factor” refers in particular to human TNF α . TNF α is found as a soluble protein as well as a precursor form called transmembrane TNF α that is expressed as a cell surface type II polypeptide. Transmembrane TNF α is processed by metalloproteinases such as TNF α -converting enzyme (TACE) between residues Ala76 and Val77, resulting in the release of the soluble form of TNF α of 157 amino acid residues. Soluble TNF α is a homotrimer of 17-kDa cleaved monomers. Transmembrane TNF α also exists as a homotrimer of 26-kD uncleaved monomers. The term “TNF α ” as used herein encompasses both the soluble and the transmembrane forms. The term “TNF α ” refers in particular to human transmembrane TNF α with UniProt ID number P01375 reproduced herein as SEQ ID NO: 134. The term “TNF α ” refers in particular to soluble transmembrane TNF α with UniProt ID number P01375 reproduced herein as SEQ ID NO: 135.

Suitably, the antibody of the disclosure is an isolated antibody. The term “isolated antibody”, as used herein, refers to an antibody that is substantially free of other antibodies having different antigenic specificities (e.g., an isolated antibody that specifically binds only IL-17A and TNF α is substantially free of antibodies that specifically bind antigens other than IL-17A and TNF α). An isolated antibody that specifically binds IL-17A and TNF α may, however, have cross-reactivity to other antigens, such IL-17A and TNF α molecules from other species. Moreover, an isolated antibody may be substantially free of other cellular material and/or chemicals.

Suitably, the antibody of the disclosure is a monoclonal antibody. The term “monoclonal antibody” or “monoclonal antibody composition” as used herein refers to antibodies that are substantially identical to amino acid sequence or are derived from the same genetic source. A monoclonal antibody composition displays a binding specificity and affinity for a particular epitope, or binding specificities and affinities for specific epitopes.

Antibodies of the disclosure include, but are not limited to, the chimeric, and humanized.

The term “chimeric antibody” is an antibody molecule in which (a) the constant region, or a portion thereof, is altered, replaced or exchanged so that the antigen binding site (variable

region) is linked to a constant region of a different or altered class, effector function and/or species, or an entirely different molecule which confers new properties to the chimeric antibody, e.g., an enzyme, toxin, hormone, growth factor, drug, etc.; or (b) the variable region, or a portion thereof, is altered, replaced or exchanged with a variable region having a different or altered antigen specificity. For example, a mouse antibody can be modified by replacing its constant region with the constant region from a human immunoglobulin. Due to the replacement with a human constant region, the chimeric antibody can retain its specificity in recognizing the antigen while having reduced antigenicity in human as compared to the original mouse antibody.

A “humanized” antibody, as used herein, is an antibody that retains the reactivity of a non-human antibody while being less immunogenic in humans. This can be achieved, for instance, by retaining the non-human CDRs and replacing the remaining parts of the antibody with their human counterparts (i.e., the constant region as well as the framework portions of the variable region). Additional framework region modifications may be made within the human framework sequences as well as within the CDR sequences derived from the germline of another mammalian species. The humanized antibodies of the disclosure may include amino acid residues not encoded by human sequences (e.g., mutations introduced by random or site-specific mutagenesis in vitro or by somatic mutation in vivo, or a conservative substitution to promote stability or manufacturing). See, e.g., Morrison et al., *Proc. Natl. Acad. Sci. USA*, 81:6851-6855, 1984; Morrison and Oi, *Adv. Immunol.*, 44:65-92, 1988; Verhoeven et al., *Science*, 239: 1534-1536, 1988; Padlan, *Molec. Immun.*, 28:489-498, 1991; and Padlan, *Molec. Immun.*, 31: 169-217, 1994. Other examples of human engineering technology include, but is not limited to Xoma technology disclosed in U.S. Pat. No. 5,766,886.

The term “recombinant humanized antibody”, as used herein, includes all humanized antibodies of the disclosure that are prepared, expressed, created or isolated by recombinant means, such as antibodies isolated from an animal (e.g. a rabbit); antibodies expressed using a recombinant expression vector transfected into a host cell, antibodies isolated from a recombinant, combinatorial human antibody library, or antibodies prepared, expressed, created or isolated by any other means that involves splicing of human immunoglobulin gene sequences to other DNA sequences. Such recombinant human antibodies have variable and constant regions (if present) derived from human germline immunoglobulin sequence. Such

antibodies can, however, be subjected to *in vitro* mutagenesis (or, when an animal transgenic for human Ig sequences is used, *in vivo* somatic mutagenesis) and thus the amino acid sequences of the VH (antibody heavy chain variable region) and VL (antibody light chain variable region) of the recombinant antibodies are sequences that, while derived from and
5 related to human germline VH and VL sequences, may not naturally exist within the human antibody germline.

Suitably, the antibody of the disclosure or a binding domain thereof is humanized.
Suitably, the antibody of the disclosure or a binding domain thereof is humanized and comprises rabbit-derived CDRs.

10 The term “multispecific antibody” as used herein, refers to an antibody that binds to two or more different epitopes on at least two or more different targets (e.g., IL-17A and TNF α), or binds to two or more different epitopes of the same target. The “multispecific antibody” of the disclosure has two or more binding domains, for example two or three binding domains. The term “multispecific antibody” includes bispecific, trispecific, tetraspecific, pentaspecific
15 and hexaspecific. The term “bispecific antibody” as used herein, refers to an antibody that binds to two different epitopes, e.g., on two different targets (e.g., IL-17A and TNF α), or on the same target. The term “trispecific antibody” as used herein, refers to an antibody that binds to three different epitopes, e.g., on three different targets (e.g., IL-17A, TNF α and HSA), or on the same target.

20 The term “multivalent antibody” refers to a single binding molecule with more than one valency, where “valency” is described as the number of antigen-binding moieties that binds to epitopes on identical target molecules. “Valent” refers to the presence of a specified number of binding domains specific for an antigen in a molecule. As such, the terms “monovalent”, “bivalent”, “tetravalent”, and “hexavalent” refer to the presence of one, two, four and six
25 binding domains, respectively, specific for an antigen in a molecule. The term “monovalent antibody”, as used herein, refers to an antibody that has a single antigen-binding moiety that binds to a single epitope on a target molecule, such as IL-17A or TNF α . The term “bivalent antibody” as used herein, refers to an antibody that has two antigen binding moieties, each of which binds to an identical epitope.

The multispecific antibody of the present disclosure may be monovalent or multivalent, e.g., bivalent, trivalent or tetravalent, preferably, monovalent, for binding to IL-17A.

The multispecific antibody of the present disclosure may be monovalent or multivalent, e.g., bivalent, trivalent or tetravalent, preferably, monovalent, for binding to TNF α . Since
5 TNF α forms a trimer, it is, potentially, trivalent and can form three dimensional immune complexes with an antibody having several domains specifically binding to TNF α , e.g., a bivalent, trivalent or multivalent antibody for binding to TNF α . To illustrate, a study of the size of immune complex formed between TNF and infliximab (a chimeric TNF α IgG antibody) and etanercept (a TNFR2 dimeric fusion protein with an IgG1 Fc), at differing
10 antigen/antibody ratios, showed that each antibody generated immune complexes with a unique size profile (Kim MS, et al., J Mol Biol. 2007;374:1374–1388). Thus, potential high immunogenicity is one of the concerns for therapeutic antibodies targeting TNF α . Hence, in a preferred embodiment, the multispecific antibody of the present disclosure is monovalent for binding to TNF α .

15 Suitably, the multispecific antibody of the disclosure comprises only one domain specifically binding IL-17A and/or only one domain specifically binding TNF α . In a preferred embodiment, the multispecific antibody of the disclosure comprises only one domain specifically binding TNF α . Suitably, the multispecific antibody of the disclosure comprises only one domain specifically binding IL-17A and only one domain specifically binding
20 TNF α . In a specific embodiment, the multispecific antibody of the present disclosure consists of a first domain specifically binding IL-17A, a second domain specifically binding TNF α , and, optionally, a polypeptide linker between the two domains. In a particular embodiment, the optional polypeptide linker is present and consists of a polypeptide having from 4 to 25 amino acid residues.

25 Suitably, the multispecific antibody of the present disclosure is advantageously capable of neutralizing the biological activity of human TNF α and human IL-17A. It will be appreciated that the term “neutralizing” as used herein refers to a reduction in biological signaling activity which may be partial or complete. Suitable assays for determining neutralization are known in the art and certain of such assays are provided in the Examples
30 herein.

In one embodiment, the antibody of the present disclosure or the first domain thereof selectively binds to human IL-17A over human IL-17B, IL-17C, IL-17D, IL-17E and IL-17F, in particular as measured by ELISA. As used herein, the terms “selectively binds to” shall mean that the antibody, composition, formulation, etc. does not significantly bind to IL-17B/C/D/E/F, but does bind to IL-17A. Selective binding is characterized by a high affinity (or low K_D) and a low to moderate IC_{50} as distinguished from nonspecific binding which usually has a low affinity (or high K_D) with a moderate to high IC_{50} . Typically, binding is considered selective when the antibody binds with a K_D of less than 10^{-7} M. Suitably, the antibody of the present disclosure or the first domain thereof binds to human IL-17A with a higher affinity or with a lower K_D than it binds to human IL-17B, IL-17C, IL-17D, IL-17E and IL-17F as measured by SPR. Suitably, the antibody of the present disclosure or the first domain thereof has IC_{50} values to IL-17B, IL-17C, IL-17D, IL-17E, and IL-17F of at least 100x greater, e.g., at least 200x greater, at least 300x greater, at least 400x greater, than the IC_{50} to IL-17A as measured by ELISA. In one embodiment, the antibody of the present disclosure or the first domain thereof binds to human IL-17A, but does not bind to human IL-17B, IL-17C, IL-17D, IL-17E and IL-17F, in particular as measured by SPR and/or ELISA.

In a further embodiment, the multispecific antibody of the disclosure further comprises a third domain having a specificity against a different antigen than IL-17A and $TNF\alpha$. Suitably, the multispecific antibody of the disclosure is a trispecific antibody. “Trispecific antibody” as used herein refers to an antibody molecule with three antigen binding domains, for example, wherein one binding domain binds human $TNF\alpha$, another binding domain binds human IL-17A, and yet another binding domain binds an antigen capable of extending the half-life of the antibody molecule, e.g., human serum albumin.

In particular, the multispecific antibody of the disclosure further comprises a third domain specifically binding to human serum albumin (HSA).

The inventors have surprisingly found that an addition of a third domain specifically binding to human serum albumin to the multispecific antibody of the disclosure comprising a first domain specifically binding IL-17A and a second domain specifically binding to $TNF\alpha$ has the following beneficial effects:

(i) increased serum half-life of the multispecific antibody of the disclosure comprising at least one domain specifically binding to human serum albumin; and

(ii) addition of a human serum albumin binding domain to the multispecific antibody of the disclosure is compatible with the functionalities of other binding domains, e.g., neutralizing activity of IL-17A and TNF α binding domains.

The term "HSA" refers in particular to human serum albumin with UniProt ID number P02768. Human Serum Albumin (HSA) is a 66.4 kDa abundant protein in human serum (50% of total protein) composed of 585 amino acids (Sugio, Protein Eng, Vol. 12, 1999, 439-446). Multifunctional HSA protein is associated with its structure that allowed binding and transporting a number of metabolites such as fatty acids, metal ions, bilirubin and some drugs (Fanali, Molecular Aspects of Medicine, Vol. 33, 2012, 209-290). HSA concentration in serum is around 3.5-5 g/dL. Albumin binding antibodies may be used for example, for extending the in vivo serum half-life of drugs or proteins conjugated thereto.

Suitably, in one embodiment the multispecific antibody of the present disclosure comprises a first domain specifically binding IL-17A and a second domain specifically binding TNF α , and a third domain specifically binding human serum albumin. The multispecific antibody of the present disclosure may be monovalent or multivalent, e.g., bivalent, trivalent or tetravalent, preferably monovalent, for binding to human serum albumin. Suitably, the multispecific antibody of the disclosure comprises only one domain specifically binding IL-17A and only one domain specifically binding TNF α , and only one domain specifically binding human serum albumin. In a specific embodiment, the multispecific antibody of the present disclosure consists of a first domain specifically binding IL-17A, a second domain specifically binding TNF α , a third domain specifically binding human serum albumin, and, optionally, a polypeptide linker between the two domains. In a particular embodiment, the optional polypeptide linker is present and consists of a polypeptide having from 4 to 25 amino acid residues.

Advantageously, the multispecific antibody of the present disclosure comprises a first domain specifically binding IL-17A and a second domain specifically binding TNF α , and, optionally, a third domain specifically binding human serum albumin, wherein said domains are capable of binding to their respective antigen(s) or receptor(s) simultaneously.

The domains of the multispecific antibody of the present disclosure, e.g., said first domain, said second domain, said third domain, are independently selected from the group consisting of a Fab, an Fv, an scFv, a dsFv, an scAb, STAB, a single domain antibody (sdAb or dAb), a single domain heavy chain antibody, and a single domain light chain antibody, a VHH, a VNAR, single domain antibodies based on the VNAR structure from shark, and binding domains based on alternative scaffolds including but limited to ankyrin-based domains, fynomers, avimers, anticalins, fibronectins, and binding sites being built into constant regions of antibodies (e.g. F-star's Modular Antibody TechnologyTM), preferably from the group consisting of a Fab, Fv and an scFv, more preferably wherein said first domain and/or said second domain and/or said third domain is/are Fv or scFv.

The multispecific antibody of the present disclosure can be in any suitable format. In one embodiment, the multispecific antibody of the disclosure is in a format selected from the group consisting of a single-chain diabody (scDb), a tandem scDb (Tandab), a linear dimeric scDb (LD-scDb), a circular dimeric scDb (CD-scDb), a bispecific T-cell engager (BiTE; tandem di-scFv), a tandem tri-scFv, a tribody (Fab-(scFv)₂) or bibody (Fab-(scFv)₁), Fab, Fab-Fv₂, Morrison (IgG CH3-scFv fusion (Morrison L) or IgG CL-scFv fusion (Morrison H)), triabody, scDb-scFv, bispecific Fab₂, di-miniantibody, tetrabody, scFv-Fc-scFv fusion, scFv-HSA-scFv fusion, di-diabody, DVD-Ig, COVD, IgG-scFab, scFab-dsscFv, Fv₂-Fc, IgG-scFv fusions, such as bsAb (scFv linked to C-terminus of light chain), Bs1Ab (scFv linked to N-terminus of light chain), Bs2Ab (scFv linked to N-terminus of heavy chain), Bs3Ab (scFv linked to C-terminus of heavy chain), Ts1Ab (scFv linked to N-terminus of both heavy chain and light chain), Ts2Ab (dsscFv linked to C-terminus of heavy chain), Bispecific antibodies based on heterodimeric Fc domains, such as Knob-into-Hole antibodies (KiHs); an Fv, scFv, scDb, tandem-di-scFv, tandem tri-scFv, Fab-(scFv)₂, Fab-(scFv)₁, Fab, Fab-Fv₂, COVD fused to the N- and/or the C-terminus of either chain of a heterodimeric Fc domain or any other heterodimerization domain, a MATCH and DuoBodies, preferably is tribody or scDb-scFv.

The term "diabodies" refers to antibody fragments with two antigen-binding sites, which fragments comprise a VH connected to VL in the same polypeptide chain (VH-VL). By using a linker that is too short to allow pairing between the two domains on the same chain, the domains are forced to pair with the complementary domains of another chain to create two antigen-binding sites. Diabodies may be bivalent or bispecific. Diabodies are described more fully in, for example, EP404097, WO 93/01161, Hudson et al., Nat. Med. 9:129-134 (2003),

and Holliger et al., Proc. Natl. Acad. Sci. U.S.A. 90: 6444-6448 (1993). Triabodies and tetraabodies are also described in Hudson et al., Nat. Med. 9:129-134 (2003). The bispecific scDb, in particular the bispecific monomeric scDb, particularly comprises two variable heavy chain domains (VH) or fragments thereof and two variable light chain domains (VL) or fragments thereof connected by linkers L1, L2 and L3 in the order VHA-L1-VLB-L2-VHB-L3-VLA, VHA-L1-VHB-L2-VLB-L3-VLA, VLA-L1-VLB-L2-VHB-L3-VHA, VLA-L1-VHB-L2-VLB-L3-VHA, VHB-L1-VLA-L2-VHA-L3-VLB, VHB-L1-VHA-L2-VLA-L3-VLB, VLB-L1-VLA-L2-VHA-L3-VHB or VLB-L1-VHA-L2-VLA-L3-VHB, wherein the VLA and VHA domains jointly form the antigen binding site for the first antigen, and VLB and VHB jointly form the antigen binding site for the second antigen. The linker L1 particularly is a peptide of 2-10 amino acids, more particularly 3-7 amino acids, and most particularly 5 amino acids, and linker L3 particularly is a peptide of 1-10 amino acids, more particularly 2-7 amino acids, and most particularly 5 amino acids. The middle linker L2 particularly is a peptide of 10-40 amino acids, more particularly 15-30 amino acids, and most particularly 20-25 amino acids.

In one embodiment, the multispecific antibody of the disclosure comprises an immunoglobulin Fc region polypeptide. The term "Fc region" herein is used to define a C-terminal region of an immunoglobulin heavy chain, including native-sequence Fc regions and variant Fc regions. Suitable native-sequence Fc regions include human IgG1, IgG2 (IgG2A, IgG2B), IgG3 and IgG4. "Fc receptor" or "FcR" describes a receptor that binds to the Fc region of an antibody. The preferred FcR is a native sequence human FcR. Moreover, a preferred FcR is one which binds an IgG antibody (a gamma receptor) and includes receptors of the FcγRI, FcγRII, and FcγRIII subclasses, including allelic variants and alternatively spliced forms of these receptors, FcγRII receptors include FcγRIIA (an "activating receptor") and FcγRIIB (an "inhibiting receptor"), which have similar amino acid sequences that differ primarily in the cytoplasmic domains thereof. Activating receptor FcγRIIA contains an immunoreceptor tyrosine-based activation motif (ITAM) in its cytoplasmic domain. Inhibiting receptor FcγRIIB contains an immunoreceptor tyrosine-based inhibition motif (ITIM) in its cytoplasmic domain, (see M. Daeron, Annu. Rev. Immunol. 5:203-234 (1997). FcRs are reviewed in Ravetch and Kinet, Annu. Rev. Immunol. 9: 457-92 (1991); Capet et al, Immunomethods 4: 25-34 (1994); and de Haas et al, J. Lab. Clin. Med. 126: 330-41 (1995). Other FcRs, including those to be identified in the future, are encompassed by the term "FcR"

herein. The term “Fc receptor” or “FcR” also includes the neonatal receptor, FcRn, which is responsible for the transfer of maternal IgGs to the fetus. Guyer et al, J. Immunol. 117: 587 (1976) and Kim et al., J. Immunol. 24: 249 (1994). Methods of measuring binding to FcRn are known (see, e.g., Ghetie and Ward, Immunol. Today 18: (12): 592-8 (1997); Ghetie et al, Nature Biotechnology 15 (7): 637-40 (1997); Hinton et al, J. Biol. Chem. TJI (8): 6213-6 (2004); WO 2004/92219 (Hinton et al). Binding to FcRn in vivo and serum half-life of human FcRn high-affinity binding polypeptides can be assayed, e.g., in transgenic mice or transfected human cell lines expressing human FcRn, or in primates to which the polypeptides having a variant Fc region are administered. WO 2004/42072 (Presta) describes antibody variants which improved or diminished binding to FcRs. See also, e.g., Shields et al, J. Biol. Chem. 9(2): 6591-6604 (2001).

In order to increase the number of specificities/functionalities at the same or lower molecular weight, it is advantageous to use antibodies comprising antibody fragments, such as Fv, Fab, Fab' and F(ab')₂ fragments and other antibody fragments. These smaller molecules retain the antigen binding activity of the whole antibody and can also exhibit improved tissue penetration and pharmacokinetic properties in comparison to the whole immunoglobulin molecules. Whilst such fragments appear to exhibit a number of advantages over whole immunoglobulins, they also suffer from an increased rate of clearance from serum since they lack the Fc domain that imparts a long half-life in vivo (Medasan et al., 1997, J. Immunol. 158:2211-2217). Molecules with lower molecular weights penetrate more efficiently into target tissues (e.g. solid cancers) and thus hold the promise for improved efficacy at the same or lower dose. Suitably, the antibody of the disclosure does not comprise an immunoglobulin Fc region polypeptide, and, optionally, does not comprise CH1 and CL regions, in particular, when said multispecific antibody comprises a third domain specifically binding human serum albumin.

Suitably, the antibody of the disclosure can be in a tribody format (Fab-(scFv)₂). Suitably, the first domain and/or the second domain and/or the third domain is/are Fab or scFv domain. In particular, the multispecific antibody of the present disclosure has one Fab domain and two scFv domains, in particular wherein scFv domains are fused to the carboxy terminus of each chain of the Fab domain. The inventors have tested the optimal relative location of the individual binding domains in trispecific formats in terms of pharmacodynamic and biophysical properties, and have surprisingly found that the multispecific antibody of the

disclosure, when in tribody format, has advantageous properties when the second domain specifically binding to TNF α is a Fab domain and the first and third domains, binding specifically to IL-17A and HSA, respectively, are scFv domains fused to said Fab domain.

Preferably, the antibody of the disclosure is in an scDb-scFv format. The term “scDb-scFv” refers to an antibody format, wherein a single-chain Fv (scFv) fragment is fused by a flexible Gly-Ser linker to a single-chain diabody (scDb). Suitably, the first domain and/or the second domain and/or the third domain is/are Fv or scFv domain. In particular, the multispecific antibody of the present disclosure, when in scDb-scFv format, has one scFv domain which is C-terminally fused to the scDb comprising of the other two domains. The multispecific antibody of the present disclosure, when in scDb-Fv format, may be represented by the formula:

VLA - L1 - VHC - L2 - VLC - L3 - VHA - L4 - VLB - L5 - VHB or
 VLB - L1 - VHA - L2 - VLA - L3 - VHB - L4 - VLC - L5 - VHC or
 VLC - L1 - VHB - L2 - VLB - L3 - VHC - L4 - VLA - L5 - VHA or
 VLA - L1 - VHB - L2 - VLB - L3 - VHA - L4 - VLC - L5 - VHC,
 preferably VLB - L1 - VHA - L2 - VLA - L3 - VHB - L4 - VLC - L5 - VHC or VLA - L1 -
 VHB - L2 - VLB - L3 - VHA - L4 - VLC - L5 - VHC,
 more preferably VLB - L1 - VHA - L2 - VLA - L3 - VHB - L4 - VLC - L5 - VHC,

wherein VLA and VHA are a light chain variable region and a heavy chain variable region of the first domain (specifically binds to IL-17A), respectively; and VLB and VHB are a light chain variable region and a heavy chain variable region of the second domain (specifically binds to TNF α), respectively; and VLC and VHC are a light chain variable region and a heavy chain variable region of the third domain (specifically binds to HSA), respectively, and wherein L1, L2, L3, L4 and L5 are polypeptide linkers.

In the context of the present disclosure, the term “polypeptide linker” refers to a linker consisting of a chain of amino acid residues linked by peptide bonds that is connecting two domains, each being attached to one end of the linker. The polypeptide linker should have a length that is adequate to link two molecules in such a way that they assume the correct conformation relative to one another so that they retain the desired activity. In particular embodiments, the polypeptide linker has a continuous chain of between 2 and 30 amino acid

residues (e.g., 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, or 30 amino acid residues). In addition, the amino acid residues selected for inclusion in the polypeptide linker should exhibit properties that do not interfere significantly with the activity of the polypeptide. Thus, the linker peptide on the whole should not exhibit a charge that would be inconsistent with the activity of the polypeptide, or interfere with internal folding, or form bonds or other interactions with amino acid residues in one or more of the monomers that would seriously impede the binding of monomer domains. In particular embodiments, the polypeptide linker is non-structured polypeptide. Useful linkers include glycine-serine, or GS linkers. By "Gly-Ser" or "GS" linkers is meant a polymer of glycines and serines in series (including, for example, (Gly-Ser)_n, (GSGGS)_n (GGGGS)_n and (GGGS)_n, where n is an integer of at least one), glycine-alanine polymers, alanine-serine polymers, and other flexible linkers such as the tether for the shaker potassium channel, and a large variety of other flexible linkers, as will be appreciated by those in the art. Glycine-serine polymers are preferred since both of these amino acids are relatively unstructured, and therefore may be able to serve as a neutral tether between components. Secondly, serine is hydrophilic and therefore able to solubilize what could be a globular glycine chain. Third, similar chains have been shown to be effective in joining subunits of recombinant proteins such as single chain antibodies.

Suitably, said L1 and L3 of the present disclosure are as set forth in SEQ ID NO: 132.
Suitably, said L2, L4 and L5 of the present disclosure are as set forth in SEQ ID NO: 23.

The inventors have tested the optimal relative location of the individual binding domains in trispecific formats in terms of pharmacodynamic and biophysical properties, and have surprisingly found that the multispecific antibody of the disclosure, when in scDb-scFv format, has advantageous properties when first domain and said second domain, binding specifically to IL-17A and TNF α , respectively, form scDb and said third domain specifically binding to HSA is scFv.

The multispecific antibody of the present disclosure has the following advantageous properties:

(a) has the ability to neutralize IL-17A with a potency relative to that of secukinumab (relative potency), determined by measuring Gro- α secretion in an HT-29 assay,

greater than 2, e.g. greater than 5, greater than 10, greater than 15, greater than 20, greater than 25, greater than 30, greater than 35, greater than 40, greater than 45, preferably greater than 50, and wherein said relative potency is the ratio of the IC₅₀ value in ng/mL of secukinumab as measured in the HT-29 assay to the IC₅₀ value in ng/mL of said multispecific antibody as measured in the HT-29 assay; and

(b) has the ability to neutralize TNF α with a potency relative to that of an scDb according to SEQ ID NO: 149 (A13) (relative potency), determined by measuring Gro- α secretion in an HT-29 assay, of at least 1, e.g., greater than 1, greater than 1.5, greater than 2, greater than 2.5, greater than 3, greater than 3.5, preferably greater than 4, more preferably greater than 4.5, and wherein said relative potency is the ratio of the IC₅₀ value in nM of said scDb according to SEQ ID NO: 149 as measured in the HT-29 assay to the IC₅₀ value in nM of said multispecific antibody as measured in the HT-29 assay; and

(c) optionally, has the ability to block interaction between IL-17A and IL-17RA with a potency relative to that of secukinumab (relative potency), determined in ELISA assay, greater than 2, e.g., greater than 3, greater than 4, greater than 5, greater than 6, greater than 7, greater than 8, greater than 9, preferably greater than 10, and wherein said relative potency is the ratio of the IC₅₀ value in ng/mL of secukinumab as measured by ELISA to the IC₅₀ value in ng/mL of said multispecific antibody as measured by ELISA; and

(d) optionally, has the ability to neutralize TNF α with a potency relative to that of an scDb according to SEQ ID NO: 149 (A13) (relative potency), determined in L929 assay, of at least 0.4, e.g., at least 0.5, preferably at least 1, and wherein said relative potency is the ratio of the IC₅₀ value in nM of said scDb according to SEQ ID NO: 149 as measured in the L929 assay to the IC₅₀ value in nM of said multispecific antibody as measured in the L929 assay; and/or

(e) binds to human IL-17A with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, less than 2 nM, than 1 nM, preferably less than 0.5 nM, as measured by surface plasmon resonance; and optionally, binds to cynomolgus IL-17A with a K_D of less than 5 nM, e.g., less than 4 nM, less than 3 nM, less than 2 nM, than 1 nM, preferably less than 0.5 nM as measured by surface plasmon resonance;

(f) binds to human TNF α with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, less than 2 nM, less than 1 nM, preferably less than 0.5 nM, more preferably less than 0.25 nM, as measured by surface plasmon resonance; and

(g) optionally, binds to human serum albumin with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, preferably less than 2 nM, as measured by surface plasmon resonance, and optionally, binds to cynomolgus serum albumin with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, preferably less than 2 nM as measured by surface plasmon resonance.

As used herein, the term “affinity” refers to the strength of interaction between antibody and antigen at single antigenic sites. Within each antigenic site, the variable region of the antibody “arm” interacts through weak non-covalent forces with antigen at numerous sites; the more interactions, the stronger the affinity.

“Binding affinity” generally refers to the strength of the sum total of non-covalent interactions between a single binding site of a molecule (e.g., of an antibody) and its binding partner (e.g., an antigen). Unless indicated otherwise, as used herein, “binding affinity”, “bind to”, “binds to” or “binding to” refers to intrinsic binding affinity that reflects a 1:1 interaction between members of a binding pair (e.g., an antibody and antigen). The affinity of a molecule X for its partner Y can generally be represented by the dissociation constant (K_D). Affinity can be measured by common methods known in the art, including those described herein. Low-affinity antibodies generally bind antigen slowly and tend to dissociate readily, whereas high-affinity antibodies generally bind antigen faster and tend to remain bound longer. A variety of methods of measuring binding affinity are known in the art, any of which can be used for purposes of the present disclosure. Specific illustrative and exemplary embodiments for measuring binding affinity, i.e. binding strength are described in the following.

The term “ k_{assoc} ”, “ k_a ” or “ k_{on} ”, as used herein, is intended to refer to the association rate of a particular antibody-antigen interaction, whereas the term “ k_{dis} ”, “ k_d ” or “ k_{off} ”, as used herein, is intended to refer to the dissociation rate of a particular antibody-antigen interaction. In one embodiment, the term “ K_D ”, as used herein, is intended to refer to the dissociation constant, which is obtained from the ratio of k_d to k_a (i.e. k_d/k_a) and is expressed as a molar concentration (M). The “ K_D ” or “ K_D value” or “ KD ” or “ KD value” according to this disclosure is in one embodiment measured by using surface-plasmon resonance assays using a MASS-1 SPR instrument (Sierra Sensors) as described in Examples. The binding affinity of an antibody may be determined, for example, by the dissociation constant (K_D). A

stronger affinity is represented by a lower K_D , while a weaker affinity is represented by a higher K_D .

Thus, in a suitable embodiment, the antibody of the disclosure binds to human IL-17A with a dissociation constant (K_D) of between 1 pM and 10 nM, 1 pM and 7 nM, 1 pM and 5 nM, 1 pM and 4 nM, 1 pM and 3 nM, 1 pM and 2.5 nM, 1 pM and 2 nM, 1 pM and 1.5 nM, 1 pM and 1 nM, preferably 1 pM and 0.5 nM, as measured by surface plasmon resonance. In a suitable embodiment, the antibody of the disclosure binds to human IL-17A with a dissociation constant (K_D) of between 1 and 500 pM as measured by surface plasmon resonance. In a suitable embodiment, the antibody of the disclosure binds to human IL-17A with a dissociation constant (K_D) of less than 5 nM, less than 4 nM, less than 3 nM, less than 2 nM, less than 1 nM, preferably less than 0.5 nM, as measured by surface plasmon resonance. Suitably, the antibody of the disclosure binds to human IL-17A with a dissociation constant (K_D) of less than 1 nM. Suitably, the antibody of the disclosure binds to human IL-17A with a dissociation constant (K_D) of less than 0.5 nM. In a further embodiment, the antibody of the disclosure binds to Cynomolgus IL-17A with a K_D of less than 10 nM, e.g. less than 7 nM, less than 5 nM, less than 4 nM, less than 3 nM, less than 2 nM, less than 1 nM, preferably less than 0.5 nM as measured by surface plasmon resonance (SPR).

Suitably, the antibody of the disclosure binds to human TNF α with a dissociation constant (K_D) of between 1 pM and 10 nM, 1 pM and 7 nM, 1 pM and 5 nM, 1 pM and 4 nM, 1 pM and 3 nM, 1 pM and 2.5 nM, 1 pM and 2 nM, 1 pM and 1.5 nM, 1 pM and 1 nM, preferably 1 pM and 0.5 nM, more preferably 1 pM and 0.25 nM, as measured by surface plasmon resonance. In a suitable embodiment, the antibody of the disclosure binds to human TNF α with a dissociation constant (K_D) of between 1 and 500 pM, preferably 1 and 250 pM, as measured by surface plasmon resonance. In a suitable embodiment, the antibody of the disclosure binds to human TNF α with a dissociation constant (K_D) of less than 5 nM, less than 4 nM, less than 3 nM, less than 2 nM, less than 1 nM, preferably less than 0.5 nM, more preferably less than 0.25 nM, as measured by surface plasmon resonance. Suitably, the antibody of the disclosure binds to human TNF α with a dissociation constant (K_D) of less than 0.5 nM. Suitably, the antibody of the disclosure binds to human TNF α with a dissociation constant (K_D) of less than 0.25 nM.

In a suitable embodiment, the antibody of the disclosure binds to human serum albumin with a dissociation constant (K_D) of between 1 pM and 10 nM, 1 pM and 7 nM, 1 pM and 5 nM, 1 pM and 4 nM, 1 pM and 3 nM, preferably 1 pM and 2 nM, as measured by surface plasmon. In a suitable embodiment, the antibody of the disclosure binds to human serum albumin with a dissociation constant (K_D) of between 1 and 2000 pM as measured by surface plasmon resonance. In a suitable embodiment, the antibody of the disclosure binds to human serum albumin with a dissociation constant (K_D) of less than 5 nM, less than 4 nM, less than 3 nM, preferably less than 2 nM, as measured by surface plasmon resonance. Suitably, the antibody of the disclosure binds to human serum albumin with a dissociation constant (K_D) of less than 2 nM. In a further embodiment, the antibody of the disclosure binds to Cynomolgus serum albumin with a K_D of less than 10 nM, e.g. less than 7 nM, less than 5 nM, than 4 nM, less than 3 nM, preferably less than 2 nM as measured by surface plasmon resonance (SPR).

Suitably, the antibody of the disclosure has beneficial biophysical properties.

- (a) has a melting temperature (T_m), determined by differential scanning fluorimetry, of at least 55°C, preferably of at least 58°C, more preferably at least 60°C in phosphate-citrate buffer at pH 6.4, 150 mM NaCl;
- (b) has a loss in monomer content, after five consecutive freeze-thaw cycles, of less than 5%, e.g., less than 4%, less than 3%, less than 2%, preferably 1% or less, when said multispecific antibody is at a starting concentration of 10 mg/ml in phosphate buffered saline (PBS), pH 7.4;
- (c) has a loss in monomer content, after storage for at least two weeks, particularly for at least four weeks, at 4°C, of less than 10%, preferably less than 5%, when said multispecific antibody is at a starting concentration of 10 mg/ml in phosphate buffered saline (PBS), pH 7.4; and/or
- (d) has a loss in monomer content, after storage for at least two weeks, particularly for at least four weeks, at 37°C, of less than 20%, preferably less than 15%, when said multispecific antibody is at a starting concentration of 10 mg/ml in phosphate buffered saline (PBS), pH 7.4.

The melting (T_m) is determined by differential scanning fluorimetry (DSF) as described earlier (Egan, et al., MAbs, 9(1) (2017), 68-84; Niesen, et al., Nature Protocols, 2(9) (2007) 2212-2221). The midpoint of transition for the thermal unfolding is determined by

Differential Scanning Fluorimetry (DSF) using the fluorescence dye SYPRO® Orange (see Wong & Raleigh, Protein Science 25 (2016) 1834-1840). Samples in phosphate-citrate buffer at pH 6.4 are prepared at a final protein concentration of 50 µg/mL and a final concentration of 5x SYPRO® Orange in a total volume of 100 µl. Twenty-five microliters of prepared
5 samples are added in triplicate to white-walled AB gene PCR plates. The assay is performed in a qPCR machine used as a thermal cycler, and the fluorescence emission is detected using the software's custom dye calibration routine. The PCR plate containing the test samples is subjected to a temperature ramp from 25°C to 96°C in increments of 1°C with 30 s pauses after each temperature increment. The total assay time is about two hours. The T_m is
10 calculated by the software GraphPad Prism using a mathematical second derivative method to calculate the inflection point of the curve. The reported T_m is an average of three measurements.

The loss in monomer content is determined by SE-HPLC. SE-HPLC is a separation technique based on a solid stationary phase and a liquid mobile phase as outlined by the USP
15 chapter 621. This method separates molecules based on their size and shape utilizing a hydrophobic stationary phase and aqueous mobile phase. The separation of molecules is occurring between the void volume (V₀) and the total permeation volume (V_T) of a specific column. Measurements by SE-HPLC are performed on a Chromaster HPLC system (Hitachi High-Technologies Corporation) equipped with automated sample injection and a UV
20 detector set to the detection wavelength of 280 nm. The equipment is controlled by the software EZChrom Elite (Agilent Technologies, Version 3.3.2 SP2) which also supports analysis of resulting chromatograms. Protein samples are cleared by centrifugation and kept at a temperature of 6°C in the autosampler prior to injection. For the analysis of scFv samples the column Shodex KW403-4F (Showa Denko Inc., # F6989202) is employed with a
25 standardized buffered saline mobile phase (50 mM sodium-phosphate pH 6.5, 300 mM sodium chloride) at the recommended flow rate of 0.35 mL/min. The target sample load per injection was 5 µg. Samples are detected by an UV detector at a wavelength of 280 nm and the data recorded by a suitable software suite. The resulting chromatograms are analyzed in the range of V₀ to V_T thereby excluding matrix associated peaks with >10 min elution time.

30 ***Exemplary domains specifically binding to IL-17A***

The multispecific antibody of the disclosure comprises a first domain specifically binding IL-17A, wherein said domain comprises a heavy chain variable region (VH) and a light chain variable region (VL), and wherein: (a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, and (b) said VL
5 comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3.

Suitable domains specifically binding IL-17A for use in the multispecific antibody of the present disclosure include, but are not limited to:

- the humanized monoclonal antibodies or binding domains thereof presented below in
10 Section “Anti-IL-17A antibodies of the disclosure”, whose sequences are listed in TABLE 1;
- AIN457 (also referred to as secukinumab; disclosed in U.S. Patent No.7, 807,155 and WO 2006/013107, which are incorporated herein by reference in their entirety) or an antigen-binding fragment thereof;
- LY2439821 (also referred to as ixekizumab; disclosed in U.S. Patent Nos. 7,838,638
15 and 8,110,191 and WO 2007/070750, which are incorporated herein by reference in their entirety) or an antigen-binding fragment thereof;
- SCH900117 or an antigen-binding fragment thereof (Merck);
- RG4943 or an antigen-binding fragment thereof (Roche);
- anti-IL-17A antibodies or antigen-binding fragments thereof are disclosed in WO
20 2006/013107, WO 2006/054059, WO 2007/070750, WO 2007/149032, WO 2008/001063, WO 2008/021156, WO 2010/034443, WO 2010/102251, WO 2012/018767, WO 2014/161570, WO 2014/001368, WO 2014/122613, WO 2015/070697, WO 2015/137843, WO 2016/048188, WO 2016/113557, WO 2016/138842, WO 2017/068472, which are incorporated herein by reference in their entirety.

25 Preferred domains specifically binding IL-17A for use in the multispecific antibody of the present disclosure include, but are not limited to, the humanized monoclonal antibodies or binding domains thereof presented below, whose sequences are listed in TABLE 1.

Thus, in one embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α ,
30 wherein said first domain comprises a set of CDRs: HCDR1, HCDR2, HCDR3, LCDR1,

LCDR2 and LCDR3, wherein the set of CDRs has 10 or fewer amino acid substitutions, e.g., 9 or fewer amino acid substitutions, 8 or fewer amino acid substitutions, 7 or fewer amino acid substitutions, 6 or fewer amino acid substitutions, 5 or fewer amino acid substitutions, 4 or fewer amino acid substitutions, 3 or fewer amino acid substitutions, 2 or fewer amino acid substitutions, 1 or 0 amino acid substitutions, preferably 0 amino acid substitutions, from a set of CDRs (i) in which HCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 1, 4, and 7, preferably SEQ ID NO: 1; HCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 2, 5, and 8, preferably SEQ ID NO: 2; HCDR3' is amino acid sequence selected from any one of SEQ ID Nos: 3, 6, and 9, preferably SEQ ID NO: 3; LCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 12, 15, and 18, preferably SEQ ID NO: 12; LCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 13, 16, and 19, preferably SEQ ID NO: 13; and LCDR3' having the amino acid sequence selected from any one of SEQ ID Nos: 14, 17, and 20, preferably SEQ ID NO: 14; or (ii) in which HCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45, preferably SEQ ID NO: 39; HCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 40, 43, and 46, preferably SEQ ID NO: 40; HCDR3' is amino acid sequence selected from any one of SEQ ID Nos: 41, 44, and 47, preferably SEQ ID NO: 41; LCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 50, 53, and 56, preferably SEQ ID NO: 50; LCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 51, 54, and 57, preferably SEQ ID NO: 51; and LCDR3' having the amino acid sequence selected from any one of SEQ ID Nos: 52, 55, and 58, preferably SEQ ID NO: 52.

In particular, the present disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises a VH CDR having an amino acid sequence of any one of the VH CDRs listed in TABLE 1. In particular, said domain comprises (or alternatively, consisting of) one, two, three, or more VH CDRs having an amino acid sequence of any of the VH CDRs listed in TABLE 1.

Suitably, the first domain specifically binding IL-17A comprises a heavy chain variable region (VH), wherein said VH comprises, in sequence, (i) the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 1, 4, and 7, said HCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 2, 5, and 8, said HCDR3 having the

amino acid sequence selected from any one of SEQ ID Nos: 3, 6, and 9. In particular, the disclosure provides antibodies that have a binding specificity for human IL-17A and comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively; or (ii) the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45, said HCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 40, 43, and 46, said HCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 41, 44, and 47. In particular, the disclosure provides antibodies that have a binding specificity for human IL-17A and comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively.

The present disclosure also provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises a VL CDR having an amino acid sequence of any one of the VL CDRs listed in TABLE 1. In particular, the first domain specifically binding IL-17A comprises (or alternatively, consisting of) one, two, three or more VL CDRs having an amino acid sequence of any of the VL CDRs listed in TABLE 1.

Suitably, said first domain specifically binding IL-17A comprises a light chain variable region (VL), wherein said VL comprises, in sequence, (i) the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 12, 15, and 18, said LCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 13, 16, and 19, said LCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 14, 17, and 20. In particular, the disclosure provides antibodies that have a binding specificity for human IL-17A and comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (ii) the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 50, 53, and 56, said LCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 51, 54, and 57, said LCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 52, 55, and 58. In particular, the disclosure provides antibodies that have a binding specificity for human IL-17A and comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively.

Suitably, the present disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises a heavy chain variable region (VH) and a light chain variable region (VL),

5 (i) wherein

(a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 1, 4, and 7, said HCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 2, 5, and 8, said HCDR3 having the amino acid sequence
10 selected from any one of SEQ ID Nos: 3, 6, and 9; and

(b) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 12, 15, and 18, said LCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 13, 16, and 19, said LCDR3 having the amino acid sequence
15 selected from any one of SEQ ID Nos: 14, 17, and 20; or

(ii) wherein

(a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45, said HCDR2 having the amino acid sequence
20 selected from any one of SEQ ID Nos: 40, 43, and 46, said HCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 41, 44, and 47; and

(b) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 50, 53, and 56, said LCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 51, 54, and 57, said LCDR3 having the amino acid sequence
25 selected from any one of SEQ ID Nos: 52, 55, and 58.

In particular, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises (i) (a) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOS:

1, 2, and 3, respectively, and (b) LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (ii) (a) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively, and (b) LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively.

5 Other domains of the disclosure specifically binding IL-17A include amino acids that have been mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in the CDR regions with the CDR regions depicted in the sequences described in TABLE 1. Suitably, other domains of the disclosure specifically binding IL-17A include mutant amino acid sequences wherein no more than 1, 2, 3, 4, 5 or 10 amino acids have been
10 mutated by amino acid deletion, insertion or substitution in the CDR regions when compared with the CDR regions depicted in the sequences described in TABLE 1. Mutations, e.g., substitutions, may potentially be made at any residue within the set of CDRs, and may be within CDR1, CDR2 and/or CDR3.

The term “amino acid” refers to naturally occurring and synthetic amino acids, as well
15 as amino acid analogs and amino acid mimetics that function in a manner similar to the naturally occurring amino acids. Naturally occurring amino acids are those encoded by the genetic code, as well as those amino acids that are later modified, e.g., hydroxyproline, gamma-carboxyglutamate, and O-phosphoserine. The terms “polypeptide” and “protein” are used interchangeably herein to refer to a polymer of amino acid residues. The terms apply to
20 amino acid polymers in which one or more amino acid residue is an artificial chemical mimetic of a corresponding naturally occurring amino acid, as well as to naturally occurring amino acid polymers and non-naturally occurring amino acid polymer. Unless otherwise indicated, a particular polypeptide sequence also implicitly encompasses conservatively modified variants thereof.

25 The techniques required to make mutations, e.g., substitutions, within amino acid sequences of CDRs, antibody VH or VL domains and antibodies generally are available in the art. Variant sequences may be made, with mutations, e.g., substitutions, that may or may not be predicted to have a minimal or beneficial effect on activity, and tested for ability to bind and/or neutralize IL-17A, or, alternatively, TNF α or human serum albumin, and/or for any
30 other desired property. Suitable mutations, e.g., substitutions, within CDRs do not result in loss of function, so an antibody of the disclosure or a binding domain thereof comprising a

thus-mutated amino acid sequence retains an ability to bind and/or neutralize IL-17A, or, alternatively, TNF α or human serum albumin. For example, it may retain the same quantitative binding and/or neutralizing ability as a domain of the disclosure in which the alteration is not made, e.g., as measured in the assays described herein. Suitably, a domain of the disclosure of antigen-binding fragment comprising a thus-mutated amino acid sequence may have an improved ability to bind and/or neutralize IL-17A, or, alternatively, TNF α or human serum albumin. Suitably, a first domain of the disclosure specifically binding IL-17A and comprising a thus-mutated amino acid sequence has the ability to neutralize IL-17A with a potency relative to that of secukinumab (relative potency), determined by measuring Gro- α secretion in an HT-29 assay, greater than 2, e.g., greater than 5, greater than 10, greater than 15, greater than 20, greater than 25, greater than 30, greater than 35, greater than 40, greater than 45, preferably greater than 50, and wherein said relative potency is the ratio of the IC₅₀ value in ng/mL of secukinumab as measured in the HT-29 assay to the IC₅₀ value in ng/mL of said multispecific antibody as measured in the HT-29 assay.

In addition, suitable mutations, e.g., substitutions, within CDRs do not result in loss in solubility, stability, and producibility in high yields of the multispecific antibody of the disclosure, so the multispecific antibody of the disclosure or a binding domain thereof comprising a thus-mutated amino acid sequence retains the biophysical characteristics. For example, it may retain the same producibility in high yields and/or stability as the multispecific antibody of the disclosure in which the alteration is not made, e.g., as measured in the assays described herein. Suitably, the multispecific antibody or a binding domain thereof comprising a thus-mutated amino acid sequence may have improved biophysical characteristics.

The terms “identical” or percent “identity”, in the context of two or more nucleic acids or polypeptide sequences, refer to two or more sequences or subsequences that are the same. “Percent (%) amino acid sequence identity” and “homology” with respect to nucleic acid, a peptide, polypeptide or antibody sequence are defined as the percentage of amino acid residues in a candidate sequence that are identical with the amino acid residues in the specific peptide or polypeptide sequence, after aligning the sequences and introducing gaps, if necessary, to achieve the maximum percent sequence identity, and not considering any conservative substitutions as part of the sequence identity. Alignment for purposes of determining percent amino acid sequence identity can be achieved in various ways that are

within the skill in the art, for instance, using publicly available computer software such as BLAST, BLAST-2 or ALIGN software. Those skilled in the art can determine appropriate parameters for measuring alignment, including any algorithms needed to achieve maximal alignment over the full length of the sequences being compared.

5 For sequence comparison, typically one sequence acts as a reference sequence, to which test sequences are compared. When using a sequence comparison algorithm, test and reference sequences are entered into a computer, subsequence coordinates are designated, if necessary, and sequence algorithm program parameters are designated. Default program parameters can be used, or alternative parameters can be designated. The sequence
10 comparison algorithm then calculates the percent sequence identities for the test sequences relative to the reference sequence, based on the program parameters.

Two examples of algorithms that are suitable for determining percent sequence identity and sequence similarity are the BLAST and BLAST 2.0 algorithms, which are described in Altschul et al., Nuc Acids Res. 25:3389-3402, 1977; and Altschul et al., J. Mol. Biol.
15 215:403-410, 1990, respectively. Software for performing BLAST analyses is publicly available through the National Center for Biotechnology Information.

The percent identity between two amino acid sequences can also be determined using the algorithm of E. Meyers and W. Miller (Comput. Appl. Biosci., 4: 11-17, 1988) which has been incorporated into the ALIGN program (version 2.0), using a PAM120 weight residue
20 table, a gap length penalty of 12 and a gap penalty of 4. In addition, the percent identity between two amino acid sequences can be determined using the Needleman and Wunsch (J. Mol. Biol. 48:444-453, 1970) algorithm which has been incorporated into the GAP program in the GCG software package (available at www.gcg.com), using either a Blossom 62 matrix or a PAM250 matrix, and a gap weight of 16, 14, 12, 10, 8, 6, or 4 and a length weight of 1, 2,
25 3, 4, 5, or 6.

Suitably, a first domain specifically binding IL-17A of the multispecific antibody of the present disclosure comprises a heavy chain variable region (VH) and a light chain variable region (VL), wherein:

(i)

- (a) said VH comprises, in sequence, the three complementary determining regions
HCDR1, HCDR2 and HCDR3,
said HCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 1, 4, and 7,
5 preferably SEQ ID NO: 1;
said HCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 2, 5, and 8,
preferably SEQ ID NO: 2;
said HCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
10 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 3, 6, and 9,
preferably SEQ ID NO: 3; and/or
- (b) said VL comprises, in sequence, the three complementary determining regions
LCDR1, LCDR2 and LCDR3,
said LCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
15 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 12, 15, and 18,
preferably SEQ ID NO: 12;
said LCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 13, 16, and 19,
preferably SEQ ID NO: 13;
20 said LCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 14, 17, and 20,
preferably SEQ ID NO: 14; or
- (ii)
- (a) said VH comprises, in sequence, the three complementary determining regions
25 HCDR1, HCDR2 and HCDR3,
said HCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 39, 42, and 45,
preferably SEQ ID NO: 39;
said HCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
30 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 40, 43, and 46,
preferably SEQ ID NO: 40;
said HCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,

94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 41, 44, and 47, preferably SEQ ID NO: 41; and/or

(b) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3,

5 said LCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 50, 53, and 56, preferably SEQ ID NO: 50;

said LCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 51, 54, and 57, preferably SEQ ID NO: 51;

10 said LCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 52, 55, and 58, preferably SEQ ID NO: 52.

Suitably, the first domain specifically binding IL-17A comprises: (i) HCDR1, HCDR2, and HCDR3 having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 1, 2, and 3, respectively, and/or LCDR1, LCDR2, and LCDR3 having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (ii) HCDR1, HCDR2, and HCDR3 having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 39, 40, and 41, respectively, and/or LCDR1, LCDR2, and LCDR3 having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 50, 51, and 52, respectively.

In a further embodiment, the first domain specifically binding IL-17A comprises a heavy chain variable region VHA and a light chain variable region VLA.

25 In the context of the present disclosure the terms “VH” (variable heavy chain or heavy chain variable region), “VL” (variable light chain or light chain variable region), “Vκ” and “Vλ” refer to families of antibody heavy and light chain sequences that are grouped according to sequence identity and homology. Methods for the determination of sequence homologies, for example by using a homology search matrix such as BLOSUM (Henikoff, S. & Henikoff, J. G., Proc. Natl. Acad. Sci. U.S.A. 89 (1992) 10915-10919), and methods for the grouping of sequences according to homologies are well known to one of ordinary skill in the art. For VH,

V κ and V λ different subfamilies can be identified, as shown, for example, in Knappik et al., J. Mol. Biol. 296 (2000) 57-86, which groups VH in VH1A, VH1B and VH2 to VH6, V κ in V κ 1 to V κ 4 and V λ in V λ 1 to V λ 3. In vivo, antibody V κ chains, V λ chains, and VH chains are the result of the random rearrangement of germline κ chain V and J segments, germline λ chain V and J segments, and heavy chain V, D and J segments, respectively. To which subfamily a given antibody variable chain belongs is determined by the corresponding V segment, and in particular by the framework regions FR1 to FR3. Thus, any VH sequence that is characterized in the present application by a particular set of framework regions HFR1 to HFR3 only, may be combined with any HFR4 sequence, for example a HFR4 sequence taken from one of the heavy chain germline J segments, or a HFR4 sequence taken from a rearranged VH sequence.

Suitably, the first domain specifically binding IL-17A of the multispecific antibody of the present disclosure comprises a heavy chain variable region VHA and wherein said VHA is VH1A, VH1B, VH3 or VH4. In one embodiment, said first domain specifically binding IL-17A of the present disclosure comprises a heavy chain variable region VHA, wherein said VHA is VH4. In a preferred embodiment, said first domain specifically binding IL-17A of the present disclosure comprises a heavy chain variable region VHA, wherein said VHA is VH3.

Suitably, the first domain specifically binding IL-17A of the multispecific antibody of the present disclosure comprises a light chain variable region VLA and wherein said VLA comprises V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 frameworks FR1 to FR3, and a framework FR4, which is selected from a V κ FR4, particularly V κ 1 FR4, V κ 3 FR4, and a V λ FR4. Suitable V λ FR4 are as set forth in SEQ ID NO: 26 to SEQ ID NO: 32. In one embodiment, said first domain specifically binding IL-17A comprises V λ FR4 comprising the amino acid sequence having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably to SEQ ID NO: 26 or SEQ ID NO: 27, more preferably to SEQ ID NO: 27. Suitably, said first domain specifically binding IL-17A comprises V λ FR4 comprising the amino acid sequence selected from any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in SEQ ID NO: 26 or 27, more preferably V λ FR4 as set forth in SEQ ID NO: 27.

Thus, in one embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises:

- (i)(a) the HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and the LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (i)(b) the HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively, and the LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively;
- (ii) a VH3 or VH4 domain, preferably VH3 domain; and
- (iii) a VL domain comprising a VL framework comprising V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 FR1 to FR3, and a framework FR4, which is selected from a V κ FR4, particularly V κ 1 FR4, V κ 3 FR4, and a V λ FR4, particularly V λ FR4 comprising the amino acid sequence having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in amino acid sequence selected from any one of SEQ ID NO: 26 to SEQ ID NO: 32, more preferably V λ FR4 as set forth in SEQ ID NO: 27.

Suitably, the present disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises a VH listed in TABLE 1. Suitably, the present disclosure also provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises (or alternatively, consisting of) a VH amino acid sequence listed in TABLE 1, wherein no more than about 20 amino acids, preferably no more than about 10 amino acids, in a framework sequence (for example, a sequence which is not a CDR) have been mutated (wherein a mutation is, as various non-limiting examples, an addition, substitution or deletion). Other domains of the disclosure specifically binding IL-17A include amino acids that have been mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in the VH regions with the VH regions depicted in the sequences described in TABLE 1.

Suitably, the present disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein

said first domain comprises a VL domain listed in TABLE 1. Suitably, the present disclosure also provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises (or alternatively, consisting of) a VL amino acid sequence listed in TABLE 1, wherein no more
5 than about 20 amino acids, preferably no more than about 10 amino acids, in a framework sequence (for example, a sequence which is not a CDR) have been mutated (wherein a mutation is, as various non-limiting examples, an addition, substitution or deletion). Other domains of the disclosure specifically binding to IL-17A include amino acids that have been mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in
10 the VL regions with the VL regions depicted in the sequences described in TABLE 1.

In one embodiment, the disclosure provides the multispecific antibody a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90
15 percent, identical to the amino acid sequence SEQ ID NO: 10 or SEQ ID NO: 11, preferably SEQ ID NO: 10, and in particular wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively. In a further embodiment, the first domain specifically binding IL-17A comprises a heavy chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ
20 ID NO: 11 and wherein said heavy chain variable region comprises Q14K, G16E, and G56A (AHo numbering).

In another embodiment, the disclosure provides the multispecific antibody a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises a heavy chain variable region comprising an amino acid sequence
25 that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 48 or SEQ ID NO: 49, preferably SEQ ID NO: 48, and in particular wherein said antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively. In a further embodiment, the disclosure provides an isolated antibody that specifically binds human IL-17A wherein said
30 antibody comprises a heavy chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 49 and wherein said heavy chain variable region comprises R20T and Q141P (AHo numbering).

In another embodiment, the disclosure provides the multispecific antibody a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain (i) comprises a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 21 or SEQ ID NO: 22, preferably SEQ ID NO: 21, and in particular wherein said antibody comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (ii) comprises a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 59 or SEQ ID NO: 60, preferably SEQ ID NO: 59, and in particular wherein said antibody comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively.

In another embodiment, the disclosure provides the multispecific antibody a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises a light chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 22 and wherein said light chain variable region comprises A51P (AHo numbering).

In a further embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain (i) comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 10; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 21; or (ii) comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 48; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 59.

Preferably, said first domain specifically binding IL-17A of the multispecific antibody of the present disclosure comprises a heavy chain variable region comprising an amino acid sequence that is (i) at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 10; and a
5 light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 21, and wherein the antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and/or LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; particularly, wherein the
10 antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (ii) at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 48; and a
15 light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 59, and wherein the antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively, and/or LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively; particularly, wherein the antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs:
20 39, 40, and 41, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively.

In a specific embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain comprises (i) a VH comprising an amino acid sequence selected
25 from the group consisting of SEQ ID NOs: 10 and 11; and/or a VL thereof comprising an amino acid sequence selected from the group consisting of SEQ ID NOs: 21 and 22, or (ii)) a VH comprising an amino acid sequence selected from the group consisting of SEQ ID NOs: 48 and 19; and/or a VL thereof comprising an amino acid sequence selected from the group consisting of SEQ ID NOs: 59 and 60.

30 In particular embodiment, the first domain specifically binding IL-17A comprises (i) a VH sequence of SEQ ID NO: 10 and a VL sequence of SEQ ID NO: 21 or (ii) a VH sequence of SEQ ID NO: 48 and a VL sequence of SEQ ID NO: 59. In yet another particular

embodiment, the first domain specifically binding IL-17A comprises (i) a VH sequence of SEQ ID NO: 11 and a VL sequence of SEQ ID NO: 22 or (ii) a VH sequence of SEQ ID NO: 49 and a VL sequence of SEQ ID NO: 60.

5 In one embodiment, a domain that specifically binds to human IL-17A is a domain that is described in TABLE 1. In one embodiment, a domain that specifically binds to human IL-17A comprises an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to (i) the amino acid sequence selected from the group consisting of SEQ ID NOs: 24 and 25, preferably SEQ ID NO: 24, or (ii) the amino acid sequence selected from the group consisting of SEQ ID NOs: 61 and 62, 10 preferably SEQ ID NO: 61. In one embodiment, a domain that specifically binds to human IL-17A is as set forth (i) in SEQ ID NO: 24 or SEQ ID NO: 25, preferably SEQ ID NO: 24, or (ii) in SEQ ID NO: 61 or SEQ ID NO: 62, preferably SEQ ID NO: 61.

Other domains of the disclosure having a binding specificity for human IL-17A include those wherein the amino acids or nucleic acids encoding the amino acids have been mutated, 15 yet have at least 60, 70, 80, 90 or 95 percent identity to the sequences described in TABLE 1. In one embodiment, it includes mutant amino acid sequences wherein no more than 1, 2, 3, 4 or 5 amino acids have been mutated in the variable regions when compared with the variable regions depicted in the sequence described in TABLE 1, while retaining substantially the same activity. The term “substantially the same activity” as used herein refers to the activity 20 as indicated by substantially the same activity being at least 50%, at least 60%, at least 70%, at least 80%, at least 90%, at least 95%, at least 98% or even at least 100% or at least 110%, or at least 120%, or at least 130%, or at least 140%, or at least 150%, or at least 160%, or at least 170%, or at least 180%, or at least 190%, e.g. up to 200% of the activity as determined for the parent antibody, e.g., the multispecific antibody of the disclosure, in particular the 25 multispecific antibody of the disclosure comprising a first domain that specifically binds to human IL-17A described in Table 1 and/or a second domain that specifically binds to human TNF α described in TABLE 1.

In yet another embodiment, the present disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically 30 binding TNF α , wherein said first domain comprises amino acid sequences that are

homologous to the sequences described in Table 1, and said first domain binds to human IL-17A, and retains the desired functional properties of those domains described in TABLE 1.

In one embodiment, a domain of the disclosure specifically binding IL-17A has a heavy chain variable region comprising HCDR1, HCDR2, and HCDR3 sequences and a light chain variable region comprising LCDR1, LCDR2, and LCDR3 sequences, wherein one or more of these CDR sequences have specified amino acid sequences based on the domains described herein or conservative modifications thereof, and wherein the domains retain the desired functional properties of the antibodies of the disclosure.

The term “conservatively modified variant” or “conservative variants” applies to both amino acid and nucleic acid sequences. With respect to particular nucleic acid sequences, conservatively modified variants refer to those nucleic acids which encode identical or essentially identical amino acid sequences, or where the nucleic acid does not encode an amino acid sequence, to essentially identical sequences. Because of the degeneracy of the genetic code, a large number of functionally identical nucleic acids encode any given protein. For instance, the codons GCA, GCC, GCG and GCU all encode the amino acid alanine. Thus, at every position where an alanine is specified by a codon, the codon can be altered to any of the corresponding codons described without altering the encoded polypeptide. Such nucleic acid variations are “silent variations”, which are one species of conservatively modified variations. Every nucleic acid sequence herein which encodes a polypeptide also describes every possible silent variation of the nucleic acid. One of skill will recognize that each codon in a nucleic acid (except AUG, which is ordinarily the only codon for methionine, and TGG, which is ordinarily the only codon for tryptophan) can be modified to yield a functionally identical molecule. Accordingly, each silent variation of a nucleic acid that encodes a polypeptide is implicit in each described sequence.

For polypeptide sequences, “conservatively modified variants” or “conservative variants” include individual substitutions, deletions or additions to a polypeptide sequence which result in the substitution of an amino acid with a chemically similar amino acid. Conservative substitution tables providing functionally similar amino acids are well known in the art. Such conservatively modified variants (i.e. having one or more “conservative modifications”) are in addition to and do not exclude polymorphic variants, interspecies homologs, and alleles of the disclosure. The following eight groups contain amino acids that

are conservative substitutions for one another: 1) Alanine (A), Glycine (G); 2) Aspartic acid (D), Glutamic acid (E); 3) Asparagine (N), Glutamine (Q); 4) Arginine (R), Lysine (K); 5) Isoleucine (I), Leucine (L), Methionine (M), Valine (V); 6) Phenylalanine (F), Tyrosine (Y), Tryptophan (W); 7) Serine (S), Threonine (T); and 8) Cysteine (C), Methionine (M) (see, e.g.,
5 Creighton, Proteins (1984)). In one embodiment, the term “conservative sequence modifications” are used to refer to amino acid modifications that do not significantly affect or alter the binding characteristics of the antibody containing the amino acid sequence.

Accordingly, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein
10 said first domain comprises (or consists of):

(i)

a heavy chain variable region (VH) comprising, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, wherein said HCDR1 is amino acid sequence SEQ ID No: 1, or a conservative variant thereof; said HCDR2 is amino acid
15 sequence SEQ ID No: 2, or a conservative variant thereof; said HCDR3 is amino acid sequence selected from any one of SEQ ID No: 3, or a conservative variant thereof; and

a light chain variable region (VL) comprising, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, wherein said LCDR1 is amino acid sequence SEQ ID No: 12, or a conservative variant thereof; said LCDR2 is amino acid
20 sequence SEQ ID No: 13, or a conservative variant thereof; said LCDR3 having the amino acid sequence SEQ ID No: 14, or a conservative variant thereof; or

(ii)

a heavy chain variable region (VH) comprising, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, wherein said HCDR1 is amino acid sequence SEQ ID No: 39, or a conservative variant thereof; said HCDR2 is amino acid
25 sequence SEQ ID No: 40, or a conservative variant thereof; said HCDR3 is amino acid sequence selected from any one of SEQ ID No: 41, or a conservative variant thereof; and

a light chain variable region (VL) comprising, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, wherein said LCDR1 is amino acid

sequence SEQ ID No: 50, or a conservative variant thereof; said LCDR2 is amino acid sequence SEQ ID No: 51, or a conservative variant thereof; said LCDR3 having the amino acid sequence SEQ ID No: 52, or a conservative variant thereof,

wherein the antibody specifically binds to human IL-17A and/or neutralize IL-17A.

5 *Anti-IL-17A antibodies of the disclosure*

The present disclosure is based on the discovery of antibody molecules that specifically bind to human IL-17A and have improved affinity, efficacy, and selectivity. Moreover, the antibody of the present disclosure has improved biophysical properties, e.g. improved solubility, developability and producibility in high with relatively low impurities (> 98%, in particular > 99% monomer as detected by SE-HPLC), and stability.

In one aspect, the disclosure provides an isolated antibody having a binding specificity for human IL-17A, comprising a set of CDRs: HCDR1, HCDR2, HCDR3, LCDR1, LCDR2 and LCDR3, wherein the set of CDRs has 10 or fewer amino acid substitutions, e.g., 9 or fewer amino acid substitutions, 8 or fewer amino acid substitutions, 7 or fewer amino acid substitutions, 6 or fewer amino acid substitutions, 5 or fewer amino acid substitutions, 4 or fewer amino acid substitutions, 3 or fewer amino acid substitutions, 2 or fewer amino acid substitutions, 1 or 0 amino acid substitutions, preferably 0 amino acid substitutions, from a set of CDRs (i) in which HCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 1, 4, and 7, preferably SEQ ID NO: 1; HCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 2, 5, and 8, preferably SEQ ID NO: 2; HCDR3' is amino acid sequence selected from any one of SEQ ID Nos: 3, 6, and 9, preferably SEQ ID NO: 3; LCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 12, 15, and 18, preferably SEQ ID NO: 12; LCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 13, 16, and 19, preferably SEQ ID NO: 13; and LCDR3' having the amino acid sequence selected from any one of SEQ ID Nos: 14, 17, and 20, preferably SEQ ID NO: 14; or (ii) in which HCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45, preferably SEQ ID NO: 39; HCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 40, 43, and 46, preferably SEQ ID NO: 40; HCDR3' is amino acid sequence selected from any one of SEQ ID Nos: 41, 44, and 47, preferably SEQ ID NO: 41; LCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 50, 53, and 56, preferably SEQ ID NO:

50; LCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 51, 54, and 57, preferably SEQ ID NO: 51; and LCDR3' having the amino acid sequence selected from any one of SEQ ID Nos: 52, 55, and 58, preferably SEQ ID NO: 52.

5 The term "IL-17A" or "IL17A" refers in particular to human IL-17A with UniProt ID number Q16552 reproduced herein as SEQ ID NO: 33. The term "cynomolgus IL-17A" or "cynomolgus monkey IL-17A" refers to *Macaca fascicularis* IL-17A with UniProt ID number G1QUS7.

10 The term "IL-17B" refers in particular to human IL-17B with UniProt ID number Q9UHF5 reproduced herein as SEQ ID NO: 34. The term "IL-17C" refers in particular to human IL-17C with UniProt ID number Q9P0M4 reproduced herein as SEQ ID NO: 35. The term "IL-17D" refers in particular to human IL-17D with UniProt ID number Q8TAD2 reproduced herein as SEQ ID NO: 36. The term "IL-17E" refers in particular to human IL-17E with UniProt ID number Q9H293 reproduced herein as SEQ ID NO: 37. The term "IL-17F" refers in particular to human IL-17F with UniProt ID number Q96PD4 reproduced
15 herein as SEQ ID NO: 38.

The term "epitope" refers to a localized region of an antigen to which an antibody can specifically bind. An epitope can be, for example, contiguous amino acids of a polypeptide, or an epitope can, for example, come together from two or more non-contiguous regions of a polypeptide or polypeptides.

20 The term "antibody" and the like, as used herein, includes: whole antibodies; any antigen-binding fragments (i.e., "antigen-binding portions") or single chains of whole antibodies; and molecules comprising antibody CDRs, VH regions or VL regions (including without limitation multispecific antibodies). A naturally occurring "whole antibody" is a glycoprotein comprising at least two heavy (H) chains and two light (L) chains inter-connected by disulfide
25 bonds. Each heavy chain is comprised of a heavy chain variable region (abbreviated herein as VH) and a heavy chain constant region. The heavy chain constant region is comprised of three domains, CH1, CH2 and CH3. Each light chain is comprised of a light chain variable region (abbreviated herein as VL) and a light chain constant region. The light chain constant region is comprised of one domain, CL. The VH and VL regions can be further subdivided into
30 regions of hypervariability, termed complementarity determining regions (CDR), interspersed

with regions that are more conserved, termed framework regions (FR). Each VH and VL is composed of three CDRs and four FRs arranged from amino terminus to carboxy terminus in the following order: FR1, CDR1, FR2, CDR2, FR3, CDR3, FR4. The variable regions of the heavy and light chains contain a binding domain that interacts with an antigen. The constant regions of the antibodies may mediate the binding of the immunoglobulin to host tissues or factors, including various cells of the immune system (e.g., effector cells) and the first component (C1q) of the classical complement system.

The term “isotype”, as used herein, refers to the antibody class (e.g., IgM, IgE, IgD, IgA, IgY, and IgG, such as IgG1 or IgG4) that is provided by the heavy chain constant region genes. Isotype also includes modifies versions of one of these classes, where modifications have been made to alter the Fc function, for example, to enhance or reduce effector functions or binding to Fc receptors. Suitably, the antibody of the disclosure is an IgG selected from the group consisting of an IgG1, an IgG2, an IgG3 and an IgG4. More suitably, the antibody of the disclosure is an IgG1 or IgG4.

The terms “antigen-binding fragment”, “antigen binding portion”, and the like, as used herein, refer to one or more fragments of an intact whole antibody that retain the ability to specifically bind to a given antigen (e.g., IL-17A). Examples of binding fragments encompassed within the term “antigen binding portion” of an antibody include a Fab fragment, a monovalent fragment consisting of the VL, VH, CL and CH1 domains; a F(ab)₂ fragment, a bivalent fragment comprising two Fab fragments linked by a disulfide bridge at the hinge region; an Fd fragment consisting of the VH and CH1 domains; an Fv fragment consisting of the VL and VH domains of a single arm of an antibody; a single domain antibody (dAb) fragment (Ward et al., 1989 Nature 341:544-546), which consists of a VH domain; an isolated complementarity determining region (CDR), dsFv, an scAb, STAB, a single domain antibody (sdAb or dAb), a single domain heavy chain antibody, and a single domain light chain antibody, a VHH, a VNAR, single domain antibodies based on the VNAR structure from shark, and binding domains based on alternative scaffolds including but limited to ankyrin-based domains, fynomers, avimers, anticalins, fibronectins, and binding sites being built into constant regions of antibodies (e.g. F-star’s Modular Antibody TechnologyTM).

The term “Complementarity Determining Regions” (“CDRs”) are amino acid sequences with boundaries determined using any of a number of well-known schemes, including those

described by Kabat et al. (1991), "Sequences of Proteins of Immunological Interest," 5th Ed. Public Health Service, National Institutes of Health, Bethesda, MD ("Kabat" numbering scheme), Al-Lazikani et al., (1997) JMB 273, 927-948 ("Chothia" numbering scheme), ImMunoGenTics (IMGT) numbering (Lefranc, M.-P., The Immunologist, 7, 132-136 (1999);

5 Lefranc, M.-P. et al., Dev. Comp. Immunol., 27, 55-77 (2003) ("IMGT" numbering scheme) and numbering scheme described in Honegger & Plückthun, J. Mol. Biol. 309 (2001) 657-670 ("Aho" numbering). For example, for classic formats, under Kabat, the CDR amino acid residues in the heavy chain variable domain (VH) are numbered 31-35 (HCDR1), 50-65 (HCDR2), and 95-102 (HCDR3); and the CDR amino acid residues in the light chain variable

10 domain (VL) are numbered 24-34 (LCDR1), 50-56 (LCDR2), and 89-97 (LCDR3). Under Chothia the CDR amino acids in the VH are numbered 26-32 (HCDR1), 52-56 (HCDR2), and 95-102 (HCDR3); and the amino acid residues in VL are numbered 24-34 (LCDR1), 50-56 (LCDR2), and 89-97 (LCDR3). By combining the CDR definitions of both Kabat and

15 102 (HCDR3) in human VH and amino acid residues 24-34 (LCDR1), 50-56 (LCDR2), and 89-97 (LCDR3) in human VL. Under IMGT the CDR amino acid residues in the VH are numbered approximately 26-35 (HCDR1), 51-57 (HCDR2) and 93-102 (HCDR3), and the CDR amino acid residues in the VL are numbered approximately 27-32 (LCDR1), 50-52 (LCDR2), and 89-97 (LCDR3) (numbering according to "Kabat"). Under IMGT, the CDRs of

20 an antibody can be determined using the program IMGT/DomainGap Align.

In the context of the present disclosure, the numbering system suggested by Honegger & Plückthun ("Aho") is used (Honegger & Plückthun, J. Mol. Biol. 309 (2001) 657-670), unless specifically mentioned otherwise. Furthermore, the following residues are defined as CDRs according to Aho numbering scheme: LCDR1 (also referred to as CDR-L1): L24-L42;

25 LCDR2 (also referred to as CDR-L2): L58-L72; LCDR3 (also referred to as CDR-L3): L107-L138; HCDR1 (also referred to as CDR-H1): H27-H42; HCDR2 (also referred to as CDR-H2): H57-H76; HCDR3 (also referred to as CDR-H3): H108-H138. For the sake of clarity, the numbering system according to Honegger & Plückthun takes the length diversity into account that is found in naturally occurring antibodies, both in the different VH and VL

30 subfamilies and, in particular, in the CDRs, and provides for gaps in the sequences. Thus, in a given antibody variable domain usually not all positions 1 to 149 will be occupied by an amino acid residue.

Preferably, the “antigen-binding region” comprises at least amino acid residues 4 to 138 of the variable light (VL) chain and 5 to 138 of the variable heavy (VH) chain (in each case numbering according to Honegger & Plückthun), more preferably amino acid residues 3 to 144 of VL and 4 to 144 of VH, and particularly preferred are the complete VL and VH chains (amino acid positions 1 to 149 of VL and 1 to 149 of VH). Antigen-binding portions can also be incorporated into single domain antibodies, maxibodies, minibodies, intrabodies, diabodies, triabodies, tetrabodies, v-NAR and bis-scFv (see, e.g., Holliger and Hudson, 2005, Nature Biotechnology, 23, 9, 1 Hel 1 36). Antigen binding portions of antibodies can be grafted into scaffolds based on polypeptides such as Fibronectin type III (Fn3) (see U.S. Pat. No. 6,703,199, which describes fibronectin polypeptide monobodies). Antigen binding portions can be incorporated into single chain molecules comprising a pair of tandem Fv segments (VH-CH1-VH-CH1) which, together with complementary light chain polypeptides, form a pair of antigen binding regions (Zapata et al., 1995 Protein Eng. 8 (10): 1057-1062; and U.S. Pat. No. 5,641,870).

The terms “domain” or “domain specifically binding X” or “binding domain”, “antigen-binding fragment thereof”, “antigen binding portion” of an antibody, and the like, as used herein, refer to one or more fragments of an intact whole antibody that retain the ability to specifically bind to a given antigen (e.g., IL-17A, TNF α , HSA). Antigen binding functions of an antibody can be performed by fragments of an intact antibody. In some embodiments, a binding domain of a multispecific antibody of the present disclosure is selected from the group consisting of a Fab fragment, a monovalent fragment consisting of the VL, VH, CL and CH1 domains; a F(ab)₂ fragment, a bivalent fragment comprising two Fab fragments linked by a disulfide bridge at the hinge region; an Fd fragment consisting of the VH and CH1 domains; an Fv fragment consisting of the VL and VH domains of a single arm of an antibody; a single domain antibody (dAb) fragment (Ward et al., 1989 Nature 341:544-546), which consists of a VH domain; an isolated complementarity determining region (CDR), dsFv, an scAb, STAB, a single domain antibody (sdAb or dAb), a single domain heavy chain antibody, and a single domain light chain antibody, a VHH, a VNAR, single domain antibodies based on the VNAR structure from shark, and binding domains based on alternative scaffolds including but limited to ankyrin-based domains, fynomers, avimers, anticalins, fibronectins, and binding sites being built into constant regions of antibodies (e.g. F-star’s Modular Antibody TechnologyTM). Suitably, a binding domain of the present

disclosure is an Fv fragment (Fv). Suitably, a binding domain of the present disclosure is a single-chain Fv fragment (scFv). Suitably, a binding domain of the present disclosure is a Fab fragment.

Preferably, the “domain” or “domain specifically binding X” or “binding domain”,
5 “antigen-binding fragment thereof”, “antigen binding portion” comprises at least amino acid residues 4 to 138 of the variable light (VL) chain and 5 to 138 of the variable heavy (VH) chain (in each case numbering according to Honegger & Plückthun), more preferably amino acid residues 3 to 144 of VL and 4 to 144 of VH, and particularly preferred are the complete VL and VH chains (amino acid positions 1 to 149 of VL and 1 to 149 of VH). Antigen-
10 binding portions can also be incorporated into single domain antibodies, maxibodies, minibodies, intrabodies, diabodies, triabodies, tetrabodies, v-NAR and bis-scFv (see, e.g., Holliger and Hudson, 2005, Nature Biotechnology, 23, 9, 1 Hel l 36). Antigen binding portions of antibodies can be grafted into scaffolds based on polypeptides such as Fibronectin type III (Fn3) (see U.S. Pat. No. 6,703,199, which describes fibronectin polypeptide
15 monobodies). Antigen binding portions can be incorporated into single chain molecules comprising a pair of tandem Fv segments (VH-CH1-VH-CH1) which, together with complementary light chain polypeptides, form a pair of antigen binding regions (Zapata et al., 1995 Protein Eng. 8 (10): 1057-1062; and U.S. Pat. No. 5,641,870).

The term “binding specificity”, or “specifically binding”, as used herein, refers to the
20 ability of an individual antibody or antibody domain to react with one antigenic determinant and not with a different antigenic determinant. As used herein, the term “specifically binds to” or is “specific for” refers to measurable and reproducible interactions such as binding between a target and an antibody, or an antibody domain, which is determinative of the presence of the target in the presence of a heterogeneous population of molecules including biological
25 molecules. For example, an antibody or an antibody domain that specifically binds to a target (which can be an epitope) is an antibody or an antibody domain that binds this target with greater affinity, avidity, more readily, and/or with greater duration than it binds to other targets. In its most general form (and when no defined reference is mentioned), “specific binding” is referring to the ability of the antibody or an antibody domain to discriminate
30 between the target of interest and an unrelated molecule, as determined, for example, in accordance with a specificity assay methods known in the art. Such methods comprise, but are not limited to Western blots, ELISA, RIA, ECL, IRMA, SPR (Surface plasmon resonance)

tests and peptide scans. For example, a standard ELISA assay can be carried out. The scoring may be carried out by standard colour development (e.g. secondary antibody with horseradish peroxidase and tetramethyl benzidine with hydrogen peroxide). The reaction in certain wells is scored by the optical density, for example, at 450 nm. Typical background (= negative
5 reaction) may be about 0.1 OD; typical positive reaction may be about 1 OD. This means the ratio between a positive and a negative score can be 10-fold or higher. In a further example, an SPR assay can be carried out, wherein at least 10-fold, preferably at least 100-fold difference between a background and signal indicates on specific binding. Typically, determination of binding specificity is performed by using not a single reference molecule,
10 but a set of about three to five unrelated molecules, such as milk powder, transferrin or the like. Particular antibodies or antibody domains of the disclosure have a binding specificity for human IL-17A or for human TNF α .

The multispecific antibody of the disclosure comprises a first domain specifically binding IL-17A and a second domain specifically binding TNF α , and thus has a binding
15 specificity to IL-17A and TNF α , in particular has a binding specificity for human IL-17A and human TNF α . In one embodiment, the antibody of the disclosure has a binding specificity for human IL-17A and *Macaca fascicularis* (also known as *Cynomolgus* monkey or “*Cynomolgus*”) IL-17A. In one embodiment, the antibody of the disclosure has a binding specificity for human TNF α and *Macaca fascicularis* (also known as *Cynomolgus* monkey or
20 “*Cynomolgus*”) TNF α .

In another aspect, the disclosure relates to an antibody or an antibody domain that has a binding specificity for human IL-17A and *Macaca fascicularis* (also known as *Cynomolgus* monkey or “*Cynomolgus*”) IL-17A.

Suitably, the anti-IL-17A antibody of the disclosure is an isolated antibody.

25 Suitably, the anti-IL-17A antibody of the disclosure is a monoclonal antibody.

Anti-IL-17A antibodies of the disclosure include, but are not limited to, chimeric, and humanized antibodies.

Suitably, the anti-IL-17A antibody of the disclosure is humanized. Suitably, the anti-IL-17A antibody of the disclosure is humanized and comprises rabbit-derived CDRs.

Antibodies of the disclosure include, but are not limited to, the humanized monoclonal antibodies isolated as described herein, including in the Examples. Examples of such anti-human IL-17A antibodies are antibodies whose sequences are listed in TABLE 1. Additional details regarding the generation and characterization of the antibodies described herein are
5 provided in the Examples.

The isolated antibody of the disclosure having a binding specificity for human IL-17A comprises a heavy chain variable region (VH) and a light chain variable region (VL), wherein: (a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, and (b) said VL comprises, in sequence, the three
10 complementary determining regions LCDR1, LCDR2 and LCDR3.

The present disclosure provides antibodies that specifically bind to IL-17A protein, said antibodies comprising a VH CDR having an amino acid sequence of any one of the VH CDRs listed in TABLE 1. In particular, the disclosure provides antibodies that specifically bind to IL-17A protein, said antibodies comprising one, two, three, or more VH CDRs having an
15 amino acid sequence of any of the VH CDRs listed in TABLE 1.

The present disclosure provides an antibody having a binding specificity for human IL-17A, which comprises a heavy chain variable region (VH), wherein said VH comprises, in sequence, (i) the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 1, 4, and
20 7, said HCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 2, 5, and 8, said HCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 3, 6, and 9. In particular, the disclosure provides antibodies that have a binding specificity for human IL-17A and comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively; or (ii) the three complementary determining regions HCDR1, HCDR2
25 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45, said HCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 40, 43, and 46, said HCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 41, 44, and 47. In particular, the disclosure provides antibodies that have a binding specificity for human IL-17A and comprises HCDR1, HCDR2, and HCDR3
30 sequences of SEQ ID NOs: 39, 40, and 41, respectively.

The present disclosure also provides antibodies that specifically bind to IL-17A protein, said antibodies comprising a VL CDR having an amino acid sequence of any one of the VL CDRs listed in TABLE 1. In particular, the disclosure provides antibodies that specifically bind to IL-17A protein, said antibodies comprising one, two, three or more VL CDRs having
5 an amino acid sequence of any of the VL CDRs listed in TABLE 1.

The present disclosure provides an antibody having a binding specificity for human IL-17A, which comprises a light chain variable region (VL), wherein said VL comprises, in sequence, (i) the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 12, 15,
10 and 18, said LCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 13, 16, and 19, said LCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 14, 17, and 20. In particular, the disclosure provides antibodies that have a binding specificity for human IL-17A and comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (ii) the three complementary determining regions
15 LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 50, 53, and 56, said LCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 51, 54, and 57, said LCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 52, 55, and 58. In particular, the disclosure provides antibodies that have a binding specificity for human IL-17A and comprises LCDR1, LCDR2,
20 and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively.

Suitably, the present disclosure provides an antibody having a binding specificity for human IL-17A, which comprises a heavy chain variable region (VH) and a light chain variable region (VL),

(i) wherein

25 (a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 1, 4, and 7, said HCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 2, 5, and 8, said HCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 3, 6, and 9; and

(b)said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 12, 15, and 18, said LCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 13, 16, and 19, said LCDR3 having the amino acid sequence
5 selected from any one of SEQ ID Nos: 14, 17, and 20; or

(ii) wherein

(a)said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45, said HCDR2 having the amino acid sequence
10 selected from any one of SEQ ID Nos: 40, 43, and 46, said HCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 41, 44, and 47; and

(b)said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 50, 53, and 56, said LCDR2 having the amino acid sequence selected
15 from any one of SEQ ID Nos: 51, 54, and 57, said LCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 52, 55, and 58.

In particular, the disclosure provides antibodies that have a binding specificity for human IL-17A and comprises (i) (a) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and (b) LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (ii) (a) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39,
20 40, and 41, respectively, and (b) LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively.

Other antibodies of the disclosure include amino acids that have been mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in the CDR regions
25 with the CDR regions depicted in the sequences described in TABLE 1. Suitably, other antibodies of the disclosure includes mutant amino acid sequences wherein no more than 1, 2, 3, 4, 5 or 10 amino acids have been mutated by amino acid deletion, insertion or substitution in the CDR regions when compared with the CDR regions depicted in the sequences described in TABLE 1. Mutations, e.g., substitutions, may potentially be made at any residue
30 within the set of CDRs, and may be within CDR1, CDR2 and/or CDR3.

Suitably, an antibody of the disclosure comprising a mutated amino acid sequence is capable of inhibiting the activity of 1 ng human IL-17A at a concentration of 50 ng/ml, preferably 20 ng/ml, preferably 10 ng/ml, preferably 5 ng/ml more preferably 1 ng/ml, more preferably 0.5 ng/ml, even more preferably 0.2 ng/ml or less of said antibody by 50%, said
5 inhibitory activity is determined by measuring GRO- α secretion induced by human IL-17A in HT-29 assay in the presence of 50 pg/ml TNF α .

Suitably, the isolated antibody of the disclosure having a binding specificity for human IL-17A comprises: a heavy chain variable region (VH) and a light chain variable region (VL), wherein:

- 10 (i)
- (a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3,
said HCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 1, 4, and 7,
15 preferably SEQ ID NO: 1;
said HCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 2, 5, and 8,
preferably SEQ ID NO: 2;
said HCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
20 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 3, 6, and 9,
preferably SEQ ID NO: 3; and/or
- (b) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3,
said LCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
25 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 12, 15, and 18,
preferably SEQ ID NO: 12;
said LCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 13, 16, and 19,
preferably SEQ ID NO: 13;
30 said LCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 14, 17, and 20,
preferably SEQ ID NO: 14; or

(ii)

(a) said VH comprises, in sequence, the three complementary determining regions

HCDR1, HCDR2 and HCDR3,

said HCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
5 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 39, 42, and 45,
preferably SEQ ID NO: 39;

said HCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 40, 43, and 46,
preferably SEQ ID NO: 40;

10 said HCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 41, 44, and 47,
preferably SEQ ID NO: 41; and/or

(b) said VL comprises, in sequence, the three complementary determining regions

LCDR1, LCDR2 and LCDR3,

15 said LCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 50, 53, and 56,
preferably SEQ ID NO: 50;

said LCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 51, 54, and 57,

20 preferably SEQ ID NO: 51;

said LCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 52, 55, and 58,
preferably SEQ ID NO: 52.

Suitably, the isolated antibody of the disclosure having a binding specificity for human
25 IL-17A comprises: (i) HCDR1, HCDR2, and HCDR3 having at least 60, 70, 80, 90, 91, 92,
93, 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 1, 2, and 3,
respectively, and/or LCDR1, LCDR2, and LCDR3 having at least 60, 70, 80, 90, 91, 92, 93,
94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 12, 13, and 14,
respectively; or (ii) .HCDR1, HCDR2, and HCDR3 having at least 60, 70, 80, 90, 91, 92, 93,
30 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 39, 40, and 41,
respectively, and/or LCDR1, LCDR2, and LCDR3 having at least 60, 70, 80, 90, 91, 92, 93,

94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 50, 51, and 52, respectively.

In a further embodiment, the disclosure provides an antibody that specifically binds human IL-17A, wherein said antibody comprises a VH domain and a VL domain.

5 Suitably, the present disclosure provides an antibody that specifically binds human IL-17A, wherein said antibody comprises a VH1A, VH1B, VH3 or VH4. In one embodiment, an isolated antibody of the present disclosure comprises VH4 domain. In a preferred embodiment, an isolated antibody of the present disclosure comprises VH3 domain.

Suitably, the present disclosure provides an isolated antibody that specifically binds
10 human IL-17A, wherein said antibody comprises V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 frameworks FR1 to FR3, and a framework FR4, which is selected from a V κ FR4, particularly V κ 1 FR4, V κ 3 FR4, and a V λ FR4. Suitable V λ FR4 are as set forth in SEQ ID NO: 26 to SEQ ID NO: 32. In one embodiment the present disclosure provides an isolated antibody that specifically binds
15 human IL-17A, wherein said antibody comprises V λ FR4 comprising the amino acid sequence having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably to SEQ ID NO: 26 or SEQ ID NO: 27, more preferably to SEQ ID NO: 27. Suitably, the present disclosure provides an isolated antibody that specifically binds human IL-17A, wherein said antibody comprises V λ
20 FR4 comprising the amino acid sequence selected from any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in SEQ ID NO: 26 or SEQ ID NO: 27, more preferably V λ FR4 as set forth in SEQ ID NO: 27.

Thus, in one embodiment, the disclosure provides an antibody comprising:

(i)(a) the HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3,
25 respectively, and the LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (i)(b) the HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively, and the LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively;

(ii) a VH3 or VH4 domain, preferably VH3 domain; and

(iii) a VL domain comprising a VL framework comprising V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 FR1 to FR3, and a framework FR4, which is selected from a V κ FR4, particularly V κ 1 FR4, V κ 3 FR4, and a V λ FR4, particularly V λ FR4 comprising the amino acid sequence having at least 60, 70, 80, 90
5 percent identity to an amino acid sequence selected from any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in amino acid sequence selected from any one of SEQ ID NO: 26 to SEQ ID NO: 32, more preferably V λ FR4 as set forth in SEQ ID NO: 27.

Suitably, the present disclosure provides an isolated antibody that specifically binds human IL-17A, wherein said antibody comprises a VH domain listed in TABLE 1.

10 Suitably, the present disclosure also provides an isolated antibody that specifically binds human IL-17A, wherein said antibody comprises a VH amino acid sequence listed in TABLE 1, wherein no more than about 10 amino acids in a framework sequence (for example, a sequence which is not a CDR) have been mutated (wherein a mutation is, as various non-limiting examples, an addition, substitution or deletion).

15 Suitably, the present disclosure also provides an isolated antibody that specifically binds human IL-17A, wherein said antibody comprises a VH amino acid sequence listed in TABLE 1, wherein no more than about 20 amino acids in a framework sequence (for example, a sequence which is not a CDR) have been mutated (wherein a mutation is, as various non-limiting examples, an addition, substitution or deletion).

20 Other antibodies of the disclosure include amino acids that have been mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in the VH regions with the VH regions depicted in the sequences described in TABLE 1.

Suitably, the present disclosure provides an isolated antibody that specifically binds human IL-17A, wherein said antibody comprises a VL domain listed in TABLE 1.

25 Suitably, the present disclosure also provides an isolated antibody that specifically binds human IL-17A, wherein said antibody comprises a VL amino acid sequence listed in TABLE 1, wherein no more than about 10 amino acids in a framework sequence (for example, a sequence which is not a CDR) have been mutated (wherein a mutation is, as various non-limiting examples, an addition, substitution or deletion).

Suitably, the present disclosure also provides an isolated antibody that specifically binds human IL-17A, wherein said antibody comprises a VL amino acid sequence listed in TABLE 1, wherein no more than about 20 amino acids in a framework sequence (for example, a sequence which is not a CDR) have been mutated (wherein a mutation is, as various non-limiting examples, an addition, substitution or deletion).

Other antibodies of the disclosure include amino acids that have been mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in the VL regions with the VL regions depicted in the sequences described in TABLE 1.

In one embodiment, the disclosure provides an isolated antibody that specifically binds human IL-17A wherein said antibody comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 10 or SEQ ID NO: 11, preferably SEQ ID NO: 10, and in particular wherein said antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively. In a further embodiment, the disclosure provides an isolated antibody that specifically binds human IL-17A wherein said antibody comprises a heavy chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 11 and wherein said heavy chain variable region comprises Q14K, G16E, and G56A (AHo numbering).

In another embodiment, the disclosure provides an isolated antibody that specifically binds human IL-17A wherein said antibody comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 48 or SEQ ID NO: 49, preferably SEQ ID NO: 48, and in particular wherein said antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively. In a further embodiment, the disclosure provides an isolated antibody that specifically binds human IL-17A wherein said antibody comprises a heavy chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 49 and wherein said heavy chain variable region comprises R20T and Q141P (AHo numbering).

In another embodiment, the disclosure provides an isolated antibody that specifically binds human IL-17A wherein said antibody (i) comprises a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 21 or SEQ ID NO: 22, preferably SEQ ID NO: 21, and in particular wherein said antibody comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (ii) comprises a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 59 or SEQ ID NO: 60, preferably SEQ ID NO: 59, and in particular wherein said antibody comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively.

In a further embodiment, the disclosure provides an isolated antibody that specifically binds human IL-17A wherein said antibody comprises a light chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 22 and wherein said light chain variable region comprises A51P (AHO numbering).

In a further embodiment, the disclosure provides an antibody that specifically binds human IL-17A wherein (i) said antibody comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 10; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 21; or (ii) said antibody comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 48; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 59.

Thus, the present disclosure provides an isolated antibody that specifically binds human IL-17A, wherein (i) said antibody comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent,

preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 10; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 21, and wherein the antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and/or LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; particularly, wherein the antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; or (ii) said antibody comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 48; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 59, and wherein the antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively, and/or LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively; particularly, wherein the antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively.

In a further embodiment, the disclosure provides an isolated antibody that specifically binds human IL-17A wherein said antibody comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 49; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 60, particularly wherein said heavy chain variable region comprises R20T and Q141P (AHO numbering).

Thus, the present disclosure provides an isolated antibody that specifically binds human IL-17A, wherein said antibody comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 49; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90,

91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 60, particularly wherein said heavy chain variable region comprises R20T and Q141P (AHO numbering), and wherein the antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively, and/or
5 LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively; particularly, wherein the antibody comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively..

In a specific embodiment, the disclosure provides an isolated antibody that specifically
10 binds human IL-17A and comprises (i) a VH comprising an amino acid sequence selected from the group consisting of SEQ ID NOs: 10 and 11; and/or a VL thereof comprising an amino acid sequence selected from the group consisting of SEQ ID NOs: 21 and 22; or (ii) a VH comprising an amino acid sequence selected from the group consisting of SEQ ID NOs: 48 and 49; and/or a VL thereof comprising an amino acid sequence selected from the group
15 consisting of SEQ ID NOs: 59 and 60. In particular embodiment, the antibody of the present disclosure comprises (i) a VH sequence of SEQ ID NO: 10 and a VL sequence of SEQ ID NO: 21; or (ii) a VH sequence of SEQ ID NO: 48 and a VL sequence of SEQ ID NO: 59. In yet another particular embodiment, the antibody of the present disclosure comprises (i) a VH sequence of SEQ ID NO: 11 and a VL sequence of SEQ ID NO: 22; or (ii) a VH sequence of
20 SEQ ID NO: 49 and a VL sequence of SEQ ID NO: 60.

In one embodiment, an antibody that specifically binds to human IL-17A is an antibody that is described in TABLE 1. In one embodiment, an antibody that specifically binds to human IL-17A comprises an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid
25 sequence selected from the group consisting of (i) SEQ ID NOs: 24 and 25, preferably SEQ ID NO: 24; or (ii) SEQ ID NOs: 61 and 62, preferably SEQ ID NO: 61. In one embodiment, an antibody that specifically binds to human IL-17A is as set forth in (i) SEQ ID NO: 24 or SEQ ID NO: 25, preferably SEQ ID NO: 24; or (ii) SEQ ID NO: 61 or SEQ ID NO: 62, preferably SEQ ID NO: 61.

30 Other antibodies of the disclosure having a binding specificity for human IL-17A include those wherein the amino acids or nucleic acids encoding the amino acids have been

mutated, yet have at least 60, 70, 80, 90 or 95 percent identity to the sequences described in TABLE 1. In one embodiment, it includes mutant amino acid sequences wherein no more than 1, 2, 3, 4 or 5 amino acids have been mutated in the variable regions when compared with the variable regions depicted in the sequence described in TABLE 1, while retaining substantially the same activity. The term “substantially the same activity” as used herein refers to the activity as indicated by substantially the same activity being at least 50%, at least 60%, at least 70%, at least 80%, at least 90%, at least 95%, at least 98% or even at least 100% or at least 110%, or at least 120%, or at least 130%, or at least 140%, or at least 150%, or at least 160%, or at least 170%, or at least 180%, or at least 190%, e.g. up to 200% of the activity as determined for the parent antibody, e.g., the antibody of the disclosure, in particular the antibody of the disclosure described in TABLE 1.

In yet another embodiment, the present disclosure provides an antibody comprising amino acid sequences that are homologous to the sequences described in TABLE 1, and said antibody binds to human IL-17A, and retains the desired functional properties of those antibodies described in TABLE 1.

In one embodiment, an antibody of the disclosure has a heavy chain variable region comprising CDR1, CDR2, and CDR3 sequences and a light chain variable region comprising CDR1, CDR2, and CDR3 sequences, wherein one or more of these CDR sequences have specified amino acid sequences based on the antibodies described herein or conservative modifications thereof, and wherein the antibodies retain the desired functional properties of the antibodies of the disclosure.

Accordingly, the disclosure provides a monoclonal antibody comprising:

(i)

a heavy chain variable region (VH) comprising, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, wherein said HCDR1 is amino acid sequence SEQ ID No: 1, or a conservative variant thereof; said HCDR2 is amino acid sequence SEQ ID No: 2, or a conservative variant thereof; said HCDR3 is amino acid sequence selected from any one of SEQ ID No: 3, or a conservative variant thereof; and

a light chain variable region (VL) comprising, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, wherein said LCDR1 is amino acid sequence SEQ ID No: 12, or a conservative variant thereof; said LCDR2 is amino acid sequence SEQ ID No: 13, or a conservative variant thereof; said LCDR3 having the amino acid sequence SEQ ID No: 14, or a conservative variant thereof; or

(ii)

a heavy chain variable region (VH) comprising, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, wherein said HCDR1 is amino acid sequence SEQ ID No: 39, or a conservative variant thereof; said HCDR2 is amino acid sequence SEQ ID No: 40, or a conservative variant thereof; said HCDR3 is amino acid sequence selected from any one of SEQ ID No: 41, or a conservative variant thereof; and

a light chain variable region (VL) comprising, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, wherein said LCDR1 is amino acid sequence SEQ ID No: 50, or a conservative variant thereof; said LCDR2 is amino acid sequence SEQ ID No: 51, or a conservative variant thereof; said LCDR3 having the amino acid sequence SEQ ID No: 52, or a conservative variant thereof,

wherein the antibody specifically binds to human IL-17A and/or neutralize IL-17A.

In one embodiment, an antibody of the disclosure is optimized for expression in a mammalian cell has a heavy chain variable region and a light chain variable region, wherein one or more of these sequences have specified amino acid sequences based on the antibodies described herein or conservative modifications thereof, and wherein the antibodies retain the desired functional properties of the antibodies of the disclosure. Accordingly, the disclosure provides a monoclonal antibody optimized for expression in a mammalian cell comprising a heavy chain variable region and a light chain variable region wherein: the heavy chain variable region comprises an amino acid sequence selected from (i) any of SEQ ID NOs: 10 and 11, and conservative modifications thereof; or (ii) any of SEQ ID NOs: 48 and 49, and conservative modifications thereof; and the light chain variable region comprises an amino acid sequence selected from (i) any of SEQ ID NOs: 21 and 22, and conservative modifications thereof; or (ii) any of SEQ ID NOs: 59 and 60, and conservative modifications thereof; wherein the antibody specifically binds to human IL-17A and/or neutralize IL-17A.

In one embodiment, an antibody of the disclosure is optimized for expression in a mammalian cell has a full length heavy chain sequence and a full length light chain sequence, wherein one or more of these sequences have specified amino acid sequences based on the antibodies described herein or conservative modifications thereof, and wherein the antibodies
5 retain the desired functional properties of the antibodies of the disclosure.

As used herein, the term, “optimized” means that a nucleotide sequence has been altered to encode an amino acid sequence using codons that are preferred in the production cell or organism, generally a eukaryotic cell, for example, a cell of Pichia, a Chinese Hamster Ovary cell (CHO) or a human cell. The optimized nucleotide sequence is engineered to retain
10 completely or as much as possible the amino acid sequence originally encoded by the starting nucleotide sequence, which is also known as the “parental” sequence. The optimized sequences herein have been engineered to have codons that are preferred in mammalian cells. However, optimized expression of these sequences in other eukaryotic cells or prokaryotic cells is also envisioned herein. The amino acid sequences encoded by optimized nucleotide
15 sequences are also referred to as optimized.

Another type of variable region modification is to mutate amino acid residues within the VH and/or VL CDR1, CDR2 and/or CDR3 regions to thereby improve one or more binding properties (e.g., affinity) of the antibody of interest, known as “affinity maturation”. Site-directed mutagenesis or PCR-mediated mutagenesis can be performed to introduce the
20 mutation(s) and the effect on antibody binding, or other functional property of interest, can be evaluated in in vitro or in vivo assays as described herein and provided in the Examples. Conservative modifications (as discussed above) can be introduced. The mutations may be amino acid substitutions, additions or deletions. Moreover, typically no more than one, two, three, four or five residues within a CDR region are altered.

25 An “affinity-matured” antibody is one with one or more alterations in one or more variable domains thereof that result in an improvement in the affinity of the antibody for antigen, compared to a parent antibody that does not possess those alteration(s). In one embodiment, an affinity-matured antibody has nanomolar or even picomolar affinities for the target antigen. Affinity-matured antibodies are produced by procedures known in the art. For
30 example, Marks et al, Bio/Technology 10:779-783 (1992) describes affinity maturation by VH- and VL-domain shuffling. Random mutagenesis of HVR and/or framework residues is

described by, for example: Barbas et al. Proc Nat. Acad. Sci U.S.A. 91:3809-3813 (1994); Schier et al. Gene 169:147-155 (1995); Jackson et al, J. Immunol. 154(7):3310- 9 (1995); and Hawkins et al, J. Mol. Biol. 226:889-896 (1992). Accordingly, the disclosure provides the antibody, wherein said antibody is affinity-matured.

5 An antibody of the disclosure further can be prepared using an antibody having one or more of the VH and/or VL sequences shown herein as starting material to engineer a modified antibody, which modified antibody may have altered properties from the starting antibody. An antibody can be engineered by modifying one or more residues within one or both variable regions (i.e., VH and/or VL), for example within one or more CDR regions and/or within one
10 or more framework regions. Additionally or alternatively, an antibody can be engineered by modifying residues within the constant region(s), for example to alter the effector function(s) of the antibody.

One type of variable region engineering that can be performed is CDR grafting. Antibodies interact with target antigens predominantly through amino acid residues that are
15 located in the six heavy and light chain complementarity determining regions (CDRs). For this reason, the amino acid sequences within CDRs are more diverse between individual antibodies than sequences outside of CDRs. Because CDR sequences are responsible for most antibody-antigen interactions, it is possible to express recombinant antibodies that mimic the properties of specific naturally occurring antibodies by constructing expression vectors that
20 include CDR sequences from the specific naturally occurring antibody grafted onto framework sequences from a different antibody with different properties (see, e.g., Riechmann, L. et al., 1998 Nature 332:323-327; Jones, P. et al., 1986 Nature 321:522- 525; Queen, C. et al., 1989 Proc. Natl. Acad. Sci. U.S.A. 86: 10029-10033; U.S. Pat. No. 5,225,539 to Winter, and U.S. Pat. Nos. 5,530,101; 5,585,089; 5,693,762 and 6,180,370 to
25 Queen et al.).

Such framework sequences can be obtained from public DNA databases or published references that include germline antibody gene sequences or rearranged antibody sequences. For example, germline DNA sequences for human heavy and light chain variable region genes can be found in the "VBase" human germline sequence database (available on the
30 Internet at www.mrc-cpe.cam.ac.uk/vbase), as well as in Kabat, E. A., et al., 1991 Sequences of Proteins of Immunological Interest, Fifth Edition, U.S. Department of Health and Human

Services, NIH Publication No. 91-3242; Tomlinson, I. M., et al., 1992 J. Mol. Biol. 227:776-798; and Cox, J. P. L. et al., 1994 Eur. J Immunol. 24:827-836; the contents of each of which are expressly incorporated herein by reference. For example, germline DNA sequences for human heavy and light chain variable region genes and rearranged antibody sequences can be
5 found in "IMGT" database (available on the Internet at www.imgt.org; see Lefranc, M.P. et al., 1999 Nucleic Acids Res. 27:209-212; the contents of each of which are expressly incorporated herein by reference).

An example of framework sequences for use in the antibodies of the disclosure are those that are structurally similar to the framework sequences used by selected antibodies of the
10 disclosure, e.g., consensus sequences and/or framework sequences used by monoclonal antibodies of the disclosure. The VH CDR1, 2 and 3 sequences, and the VL CDR1, 2 and 3 sequences, can be grafted onto framework regions that have the identical sequence as that found in the germline immunoglobulin gene from which the framework sequence derive, or the CDR sequences can be grafted onto framework regions that contain one or more
15 mutations as compared to the germline sequences. For example, it has been found that in certain instances it is beneficial to mutate residues within the framework regions to maintain or enhance the antigen binding ability of the antibody (see e.g., U.S. Pat. Nos. 5,530,101; 5,585,089; 5,693,762 and 6,180,370 to Queen et al).

A wide variety of antibody /immunoglobulin frameworks or scaffolds can be employed
20 so long as the resulting polypeptide includes at least one binding region which specifically binds to IL-17A. Such frameworks or scaffolds include the five main idiotypes of human immunoglobulins, antigen-binding fragments thereof, and include immunoglobulins of other animal species, preferably having humanized aspects.

In one aspect, the disclosure pertains to a method of generating non-immunoglobulin
25 based antibodies using non-immunoglobulin scaffolds onto which CDRs of the disclosure can be grafted. Known or future non-immunoglobulin frameworks and scaffolds may be employed, as long as they comprise a binding region specific for the target IL-17A protein. Known non-immunoglobulin frameworks or scaffolds include, but are not limited to, fibronectin (Compound Therapeutics, Inc., Waltham, Mass.), ankyrin (Molecular Partners
30 AG, Zurich, Switzerland), domain antibodies (Domantis, Ltd., Cambridge, Mass., and Ablynx nv, Zwijnaarde, Belgium), lipocalin (Pieris Proteolab AG, Freising, Germany), small modular

immuno-pharmaceuticals (Trubion Pharmaceuticals Inc., Seattle, Wash.), maxyodies (Avidia, Inc., Mountain View, Calif), Protein A (Affibody AG, Sweden), and affilin (gamma-crystallin or ubiquitin) (Scil Proteins GmbH, Halle, Germany).

The antibodies according to the disclosure have valuable properties predicted to be
5 beneficial for human patients in need of a human IL-17A targeting therapy. The antibody according to the disclosure is characterized by one or more of the following properties (as determined in Examples 1 and 2):

the antibody specifically binds to human IL-17A and:

- (a) has a binding specificity for cynomolgus monkey IL-17A;
- 10 (b) selectively binds to human IL-17A over human IL-17B, IL-17C, IL-17D, IL-17E and IL-17F as measured by ELISA;
- (c) inhibits or blocks binding between IL-17A and its receptor (IL-17RA);
- (d) reduces or neutralizes IL-17A activity;
- (e) capable of inhibiting GRO- α secretion when assessed *in vitro* in HT-29 assay;
- 15 (f) has the ability to block interaction between IL-17A and IL-17RA with a potency relative to that of secukinumab (relative potency), determined in ELISA assay, greater than 5, particularly greater than 10, greater than 15, more particularly greater than 20, and wherein said relative potency is the ratio of the IC₅₀ value in ng/mL of secukinumab as measured by ELISA to the IC₅₀ value in ng/mL of the antibody of the
20 invention in the scFv format as measured by ELISA;
- (g) has the ability to neutralize IL-17A with a potency relative to that of secukinumab (relative potency), determined by measuring GRO- α secretion in an HT-29 assay, greater than 50, particularly greater than 100, more particularly greater than 150, and wherein said relative potency is the ratio of the IC₅₀ value in ng/mL of secukinumab as
25 measured in the HT-29 assay to the IC₅₀ value in ng/mL of the antibody of the invention in the scFv format as measured in the HT-29 assay;
- (h) capable of inhibiting the activity of 1 ng human IL-17A at a concentration of 1 ng/mL or less, particularly 0.5 ng/mL or less, more particularly 0.2 ng/mL or less, by 50%,
30 said inhibitory activity is determined by measuring GRO- α secretion induced by human IL-17A in HT-29 assay in the presence of 50 pg/ml TNF α ;

- (i) binds to human IL-17A with a dissociation constant (K_D) of less than 5 nM, in particular less than 1 nM, less than 0.5 nM, less than 0.2 nM, particularly less than 100 pM, more particularly less than 50 pM, as measured by surface plasmon resonance, preferably as measured by surface plasmon resonance in a direct setup;
- 5 (j) binds to Cynomolgus IL-17A with a K_D of less than 10 nM, e.g., less than 7 nM, less than 5 nM, less than 2 nM, less than 1 nM, particularly less than 0.5 nM as measured by surface plasmon resonance, particularly as measured by surface plasmon resonance in a capture setup;
- (k) when in scFv format, has a melting temperature (T_m), determined by differential
10 scanning fluorimetry, of at least 60°C, particularly of at least 62°C, more particularly of at least 65°C, and even more particularly of at least 70°C, in particular wherein said antibody is in phosphate-citrate buffer at pH 6.4, 150 mM NaCl;
- (l) when in scFv format, has a loss in monomer content, after five consecutive freeze-thaw cycles, of less than 5%, particularly less than 3%, more particularly less than 1%,
15 when the antibody of the invention is at a starting concentration of 10 mg/ml, in particular wherein said antibody is in phosphate buffered saline (PBS), pH 7.4; and/or
- (m) when in scFv format, has a loss in monomer content, after storage for at least two weeks, particularly for at least four weeks, at 4°C, of 5% or less, in particular less than 4%, less than 3%, less than 2%, particularly less than 1%, when the antibody of the
20 disclosure is at a starting concentration of 10 mg/ml, in particular wherein said antibody is in phosphate buffered saline (PBS), pH 7.4; and/or
- (n) has a loss in monomer content, after storage for at least two weeks, particularly for at least four weeks, at 37°C, of less than 5%, when the antibody of the disclosure is at a starting concentration of 10 mg/ml.

25 In one embodiment, the antibody of the present disclosure selectively binds to human IL-17A over human IL-17B, IL-17C, IL-17D, IL-17E and IL-17F as measured by ELISA. As used herein, the terms “selectively binds to” shall mean that the antibody, composition, formulation, etc. does not significantly bind to IL-17B/C/D/E/F, but does bind to IL-17A. Selective binding is characterized by a high affinity (or low K_D) and a low to moderate IC_{50} as
30 distinguished from nonspecific binding which usually has a low affinity with a moderate to high IC_{50} . Typically, binding is considered selective when the antibody binds with a K_D of less than 10^{-7} M. Suitably, the antibody of the present disclosure binds to human IL-17A with

a higher affinity or with a lower K_D than it binds to human IL-17B, IL-17C, IL-17D, IL-17E and IL-17F as measured by SPR. Suitably, the antibody of the present disclosure has IC_{50} s to IL-17B, IL-17C, IL-17D, IL-17E, and IL-17F of more than by a factor of 100 greater, particularly more than by a factor of 200 greater, more than by a factor of 300 greater, more
5 than by a factor of 400 greater than the IC_{50} to IL-17A as measured in ELISA.

The antibodies of the disclosure specifically bind to IL-17A, wherein binding to IL-17A (a) inhibits or blocks binding between IL-17A and its receptor (IL-17RA), and (b) reduces or neutralizes IL-17A activity.

As used herein, the term “neutralizing antibody” describes an antibody that is capable of
10 neutralizing the biological signaling activity of IL-17A, for example by blocking binding of IL-17A to one or more of its receptors, in particular by blocking binding of IL-17A to IL-17RA. The antibody of the present disclosure is an IL-17A neutralizing antibody. It will be appreciated that the term “neutralizing” as used herein refers to a reduction in biological signaling activity which may be partial or complete. Neutralization of IL-17A may be
15 determined by a variety of assays, examples of which are described elsewhere herein.

Thus, the antibody of the disclosure is capable of inhibiting GRO- α secretion when assessed *in vitro* in HT-29 assay (described in Examples 1 and 2). In one embodiment, the antibody of the disclosure has the ability to neutralize IL-17A with a potency relative to that of secukinumab (relative potency), determined by measuring GRO- α secretion in an HT-29
20 assay, greater than 50, preferably greater than 100, more preferably greater than 150, and wherein said relative potency is the ratio of the IC_{50} value in ng/mL of secukinumab as measured in the HT-29 assay to the IC_{50} value in ng/mL of the antibody of the disclosure in the scFv format as measured in the HT-29 assay. In a further embodiment, the antibody of the disclosure is capable of inhibiting the activity of 1 ng human IL-17A at a concentration of 1
25 ng/mL or less, preferably 0.5 ng/mL or less, more preferably 0.2 ng/mL or less, by 50%, said inhibitory activity is determined by measuring GRO- α secretion induced by human IL-17A in HT-29 assay in the presence of 50 pg/ml TNF α .

In one embodiment, the antibody of the disclosure has the ability to block interaction between IL-17A and IL-17RA with a potency relative to that of secukinumab (relative
30 potency), determined in ELISA assay, greater than 5, preferably greater than 10, greater than

15, more particularly greater than 20, and wherein said relative potency is the ratio of the IC_{50} value in ng/mL of secukinumab as measured by ELISA to the IC_{50} value in ng/mL of the antibody of the disclosure in the scFv format as measured by ELISA.

As used herein, the term “affinity” refers to the strength of interaction between antibody
5 and antigen at single antigenic sites. Within each antigenic site, the variable region of the antibody “arm” interacts through weak non-covalent forces with antigen at numerous sites; the more interactions, the stronger the affinity.

“Binding affinity” generally refers to the strength of the sum total of non-covalent interactions between a single binding site of a molecule (e.g., of an antibody) and its binding
10 partner (e.g., an antigen). Unless indicated otherwise, as used herein, “binding affinity”, “bind to”, “binds to” or “binding to” refers to intrinsic binding affinity that reflects a 1:1 interaction between members of a binding pair (e.g., an antibody fragment and antigen). The affinity of a molecule X for its partner Y can generally be represented by the dissociation constant (K_D). Affinity can be measured by common methods known in the art, including those described
15 herein. Low-affinity antibodies generally bind antigen slowly and tend to dissociate readily, whereas high-affinity antibodies generally bind antigen faster and tend to remain bound longer. A variety of methods of measuring binding affinity are known in the art, any of which can be used for purposes of the present disclosure. Specific illustrative and exemplary embodiments for measuring binding affinity, i.e. binding strength are described in the
20 following.

The term “ k_{assoc} ”, “ k_a ” or “ k_{on} ”, as used herein, is intended to refer to the association rate of a particular antibody-antigen interaction, whereas the term “ k_{dis} ”, “ k_d ” or “ k_{off} ”, as used
herein, is intended to refer to the dissociation rate of a particular antibody- antigen interaction. In one embodiment, the term “ K_D ”, as used herein, is intended to refer to the dissociation
25 constant, which is obtained from the ratio of k_d to k_a (i.e. k_d/k_a) and is expressed as a molar concentration (M). The “ K_D ” or “ K_D value” or “ KD ” or “ KD value” according to this disclosure is in one embodiment measured by using a T200 device (Biacore, GE Healthcare) To measure affinity of the humanized scFvs to human IL-17A , biotinylated human IL-17A is captured using the Biotin-CAPture kit from Biacore. After each analyte injection cycle the
30 CAP sensor chip is regenerated and new antigen is captured. The scFvs are injected as analyte using a dose response multi-cycle kinetic assay with concentrations of the analyte ranging

from 0.35 to 90 nM diluted in running buffer. Obtained sensorgrams are fitted using the 1:1 binding model. Alternatively or in addition, affinity of the humanized scFvs can be measured and analyzed using an alternative SPR assay setup: IL-17A is immobilized on a CM5 sensor chip (GE Healthcare) by amine-coupling, and serial dilutions of scFvs ranging from 0.35 to 5 90 nM are injected over the immobilized IL-17A.

Suitably, the affinity of the antibody of the disclosure to IL-17A, may be higher than the affinity of IL-17A to IL-17RA. It will be appreciated that a higher affinity of the antibody of the disclosure as compared to the affinity of IL-17A to IL-17RA may be particularly useful for dissociating or neutralizing the pre-formed IL-17RA/IL-17A complexes. In one 10 embodiment, the antibody of the disclosure neutralizes IL-17RA/IL-17A interaction. In another embodiment, the antibody of the disclosure inhibits or blocks binding between IL-17A and its receptor (IL-17RA). In one embodiment, the antibody of the disclosure neutralizes IL-17A activity.

Suitably, the affinity of the antibody of the disclosure to IL-17A may be comparable to or 15 higher, preferably higher, than the affinity of secukinumab to IL-17A. In one embodiment, the antibody of the disclosure neutralizes IL-17A activity with potency equal to or higher, preferably higher, than secukinumab. In a further embodiment, the antibody of the disclosure neutralizes IL-17RA/IL-17A interaction with potency equal to or higher, preferably higher, than secukinumab.

20 The binding affinity of an antibody may be determined, for example, by the dissociation constant (K_D). A stronger affinity is represented by a lower K_D , while a weaker affinity is represented by a higher K_D .

Thus, in a suitable embodiment, the antibody of the disclosure may have a K_D of between 1 and 10000 pM, 1 and 7000 pM, 1 and 5000 pM, 1 and 2500 pM, 1 and 2000 pM, 1 and 25 1000 pM, 1 and 750 pM, 1 and 500 pM, 1 and 400 pM, 1 and 300 pM, 1 and 200 pM, 1 and 100 pM, 1 and 50 pM, preferably as measured by surface plasmon, more preferably as measured by surface plasmon resonance in a direct setup. In a suitable embodiment, the antibody of the disclosure has a K_D of between 1 and 200 pM, in particular of between 1 and 100 pM, as measured by surface plasmon resonance in a direct setup. In a particular 30 embodiment, the antibody of the disclosure has a K_D of between 1 and 50 pM as measured by

surface plasmon resonance in a direct setup. In a suitable embodiment, the antibody of the disclosure may have a K_D of less than 5 nM, less than 4 nM, less than 3 nM, less than 2 nM, less than 1 nM, less than 0.5 nM, less than 0.4 nM, less than 0.3 nM, less than 0.25 nM, less than 0.2 nM, less than 150 pM, less than 100 pM, or less than 50 pM, preferably as measured
 5 by surface plasmon resonance, more preferably as measured by surface plasmon resonance in a direct setup. Suitably, the antibody of the disclosure has a K_D of less than 1 nM, in particular of less than 100 pM. Suitably, the antibody of the disclosure has a K_D of less than 0.5 nM, in particular of less than 50 pM. More suitably, the antibody of the disclosure has a K_D of less than 0.2 nM, in particular of less than 50 pM as measured by surface plasmon resonance in a
 10 direct setup.

In a further embodiment, the antibody of the disclosure binds to Cynomolgus IL-17A with a K_D of less than 10 nM, e.g. less than 7 nM, less than 5 nM, less than 2 nM, less than 1 nM, particularly less than 0.5 nM as measured by surface plasmon resonance (SPR) in a capture set-up.

15 Suitably, the antibody of the disclosure binds to human IL-17A with a K_{on} rate of at least $10^3 \text{ M}^{-1}\text{s}^{-1}$ or greater, at least $10^4 \text{ M}^{-1}\text{s}^{-1}$ or greater, at least $5 \times 10^4 \text{ M}^{-1}\text{s}^{-1}$ or greater, at least $10^5 \text{ M}^{-1}\text{s}^{-1}$ or greater, at least $5 \times 10^5 \text{ M}^{-1}\text{s}^{-1}$ or greater, at least $10^6 \text{ M}^{-1}\text{s}^{-1}$ or greater as measured by surface plasmon resonance (SPR), preferably as measured by surface plasmon resonance in a direct setup. Preferably, the antibody of the disclosure has a K_{on} rate of at least
 20 $10^5 \text{ M}^{-1}\text{s}^{-1}$ or greater, particularly at least $5 \times 10^5 \text{ M}^{-1}\text{s}^{-1}$ or greater, more particularly at least $10^6 \text{ M}^{-1}\text{s}^{-1}$ or greater, as measured by SPR, preferably as measured by surface plasmon resonance in a direct setup.

Suitably, the antibody of the disclosure binds to human IL-17A with a K_{off} rate of $5 \times 10^{-3} \text{ s}^{-1}$ or less, $3 \times 10^{-3} \text{ s}^{-1}$ or less, 10^{-3} s^{-1} or less, $5 \times 10^{-4} \text{ s}^{-1}$ or less, $3 \times 10^{-4} \text{ s}^{-1}$ or less, 10^{-4} s^{-1} or
 25 less, $5 \times 10^{-5} \text{ s}^{-1}$ or less, as measured by surface plasmon resonance (SPR), preferably as measured by surface plasmon resonance in a direct setup. Preferably, the antibody of the disclosure has a K_{off} rate of 10^{-4} s^{-1} or less, in particular $5 \times 10^{-5} \text{ s}^{-1}$ or less as measured by SPR, preferably as measured by surface plasmon resonance in a direct setup.

Suitably, the antibody of the disclosure has beneficial biophysical properties.

Suitably, the antibody of the disclosure has a melting temperature (T_m) of at least 60°C, preferably of at least 62°C, more preferably at least 65°C, even more preferably at least 70°C, when expressed in the scFv (single chain variable fragment) antibody format, as determined by differential scanning fluorimetry (DSF) as described earlier (Egan, et al., MABs, 9(1) (2017), 68-84; Niesen, et al., Nature Protocols, 2(9) (2007) 2212-2221). The midpoint of transition for the thermal unfolding of the scFv constructs is determined by Differential Scanning Fluorimetry using the fluorescence dye SYPRO® Orange (see Wong & Raleigh, Protein Science 25 (2016) 1834-1840). Samples in phosphate-citrate buffer at pH 6.4 are prepared at a final protein concentration of 50 µg/mL and containing a final concentration of 5x SYPRO® Orange in a total volume of 100 µl. Twenty-five microliters of prepared samples are added in triplicate to white-walled AB gene PCR plates. The assay is performed in a qPCR machine used as a thermal cycler, and the fluorescence emission is detected using the software's custom dye calibration routine. The PCR plate containing the test samples is subjected to a temperature ramp from 25°C to 96°C in increments of 1°C with 30 s pauses after each temperature increment. The total assay time is about two hours. The T_m is calculated by the software GraphPad Prism using a mathematical second derivative method to calculate the inflection point of the curve. The reported T_m is an average of three measurements.

The antibody of the disclosure, in particular when expressed in the scFv (single chain variable fragment) antibody format, is characterized by a loss in monomer content, after five consecutive freeze-thaw cycles, of less than 5%, particularly less than 3%, more particularly less than 1%, when the antibody of the disclosure is at a starting concentration of 10 mg/ml.

After storage for at least two weeks, particularly for at least four weeks at 4°C, the antibody of the disclosure, in particular when expressed in the scFv (single chain variable fragment) antibody format, is characterized by a loss in monomer content of 5% or less, particularly less than 4%, less than 3%, less than 2%, preferably less than 1%, when the antibody of the disclosure is at a starting concentration of 10 mg/ml. The loss in monomer content is as determined by area under the curve calculation of SE-HPLC chromatograms. SE-HPLC is a separation technique based on a solid stationary phase and a liquid mobile phase as outlined by the USP chapter 621. This method separates molecules based on their size and shape utilizing a hydrophobic stationary phase and aqueous mobile phase. The separation of molecules is occurring between the void volume (V_0) and the total permeation

volume (VT) of a specific column. Measurements by SE-HPLC are performed on a Chromaster HPLC system (Hitachi High-Technologies Corporation) equipped with automated sample injection and a UV detector set to the detection wavelength of 280 nm. The equipment is controlled by the software EZChrom Elite (Agilent Technologies, Version 3.3.2 SP2) which
5 also supports analysis of resulting chromatograms. Protein samples are cleared by centrifugation and kept at a temperature of 4-6°C in the autosampler prior to injection. For the analysis of scFv samples the column Shodex KW403-4F (Showa Denko Inc., #F6989202) is employed with a standardized buffered saline mobile phase (50 mM sodium-phosphate pH 6.5, 300 mM sodium chloride) at the recommended flow rate of 0.35 mL/min. The target
10 sample load per injection was 5 µg. Samples are detected by an UV detector at a wavelength of 280 nm and the data recorded by a suitable software suite. The resulting chromatograms are analyzed in the range of V0 to VT thereby excluding matrix associated peaks with >10 min elution time.

Suitably, the isolated antibody of the present disclosure is selected from the group
15 consisting of: a monoclonal antibody, a chimeric antibody, a Fab, an Fv, an scFv, dsFv, an scAb, STAB, a single domain antibody (sdAb or dAb), a single domain heavy chain antibody, and a single domain light chain antibody, a VHH, a VNAR, single domain antibodies based on the VNAR structure from shark, and binding domains based on alternative scaffolds including but limited to ankyrin-based domains, fynomers, avimers, anticalins, fibronectins,
20 and binding sites being built into constant regions of antibodies (e.g. F-star's Modular Antibody TechnologyTM), preferably scFv.

Suitably, the antibody of the disclosure is an Fv. Suitably, the antibody of the disclosure is scFv antibody fragment. "Single-chain Fv" or "scFv" or "sFv" antibody fragments
25 comprise the VH and VL domains of an antibody, wherein these domains are present in a single polypeptide chain. Generally, the Fv polypeptide further comprises a polypeptide linker between the VH and VL domains which enables the sFv to form the desired structure for target binding. "Single-chain Fv" or "scFv" antibody fragments comprise the VH and VL domains of antibody, wherein these domains are present in a single polypeptide chain. Generally, the scFv polypeptides further comprises a polypeptide linker between the VH and
30 VL domains which enables the scFv to form the desired structure for antigen binding (see, for example, Plückthun, The pharmacology of Monoclonal Antibodies, vol. 113, Rosenberg and Moore eds., (Springer-Verlag, New York, 1994), pp. 269-315). In particular embodiments,

said functional fragment is an scFv format comprising the linker according to SEQ ID NO: 23. In one embodiment, an antibody that specifically binds to human IL-17A is an antibody that is described in TABLE 1. In one embodiment, an antibody that specifically binds to human IL-17A comprises an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 5 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence selected from (i) the group consisting of SEQ ID NOs: 24 and SEQ ID NO: 25; or (ii) the group consisting of SEQ ID NOs: 61 and SEQ ID NO: 62. In a further embodiment, the antibody of the disclosure is a single-chain variable fragment (scFv) (i) as shown in SEQ ID NO: 24 or in SEQ ID NO: 25; or (ii) as shown in SEQ ID NO: 61 or in SEQ ID NO: 62.

10 Suitably, the antibody of the disclosure is an IgG antibody. In one embodiment, the antibody of the disclosure is an IgG selected from the group consisting of an IgG1, an IgG2, an IgG3 and an IgG4, preferably an IgG1.

Exemplary domains specifically binding to TNF α

The multispecific antibody of the disclosure comprises a second domain specifically 15 binding TNF α , wherein said domain comprises a heavy chain variable region (VH) and a light chain variable region (VL), wherein: (a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, and (b) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3.

20 Suitable domains specifically binding TNF α for use in the multispecific antibody of the present disclosure include, but are not limited to:

- the humanized monoclonal antibodies or binding domains thereof whose sequences are listed in TABLE 1 (described in WO 2017/158101, which is incorporated herein by reference in its entirety);

25

- infliximab (Remicade®; U.S. Pat. Nos. 6,277,969, 6,284,471, 6,790,444, and 6,835,823, all of which are incorporated herein by reference);

- adalimumab / D2E7 (Humira®; described in U.S. Pat. No. 6,090,382, which is incorporated herein by reference in its entirety);

30

- certolizumab, a PEGylated Fab fragment (Cimzia®; described in U.S. 7,012,135 and U.S. 7,186,820, all of which are incorporated herein by reference);

• golimumab (Simponi®; published U.S. application 2009/214528, which is incorporated herein by reference in its entirety).

Preferred domains specifically binding TNF α for use in the multispecific antibody of the present disclosure include, but are not limited to, the humanized monoclonal antibodies or
5 binding domains thereof whose sequences are listed in TABLE 1 (described in WO 2017/158101, which is incorporated herein by reference in its entirety).

Thus, in one embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises a set of CDRs: HCDR1, HCDR2, HCDR3, LCDR1,
10 LCDR2 and LCDR3, wherein the set of CDRs has 10 or fewer amino acid substitutions, e.g., 9 or fewer amino acid substitutions, 8 or fewer amino acid substitutions, 7 or fewer amino acid substitutions, 6 or fewer amino acid substitutions, 5 or fewer amino acid substitutions, 4 or fewer amino acid substitutions, 3 or fewer amino acid substitutions, 2 or fewer amino acid substitutions, 1 or 0 amino acid substitutions, preferably 0 amino acid substitutions, from a set
15 of CDRs in which HCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 25, 28, and 31, preferably SEQ ID NO: 25; HCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 26, 29, and 32, preferably SEQ ID NO: 26; HCDR3' is amino acid sequence selected from any one of SEQ ID Nos: 27, 30, and 33, preferably SEQ ID NO: 27; LCDR1' is amino acid sequence selected from any one of SEQ ID Nos: 38, 41, and 44,
20 preferably SEQ ID NO: 38; LCDR2' is amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45, preferably SEQ ID NO: 39; and LCDR3' having the amino acid sequence selected from any one of SEQ ID Nos: 40, 43, and 46, preferably SEQ ID NO: 40.

In particular, the present disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α ,
25 wherein said second domain comprises a VH CDR having an amino acid sequence of any one of the VH CDRs listed in TABLE 1. In particular, the second domain specifically binding TNF α comprises (or alternatively, consisting of) one, two, three, or more VH CDRs having an amino acid sequence of any of the VH CDRs listed in TABLE 1.

Suitably, the present disclosure provides a second domain specifically binding TNF α ,
30 wherein said second domain comprises a heavy chain variable region (VH), wherein said VH

comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 25, 28, and 31, said HCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 26, 29, and 32, said HCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 27, 30, and 33. In particular, said second domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively.

The present disclosure also provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises a VL CDR having an amino acid sequence of any one of the VL CDRs listed in TABLE 1. In particular, said second domain specifically binding TNF α comprises (or alternatively, consisting of) one, two, three or more VL CDRs having an amino acid sequence of any of the VL CDRs listed in TABLE 1.

Suitably, said second domain specifically binding TNF α comprises a light chain variable region (VL), wherein said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 38, 41, and 44, said LCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45, said LCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 40, 43, and 46. In particular, said second domain comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively.

Suitably, the present disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises a heavy chain variable region (VH) and a light chain variable region (VL), wherein:

(a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 25, 28, and 31, said HCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 26, 29, and 32, said HCDR3 having the amino acid sequence selected from any one of SEQ ID Nos: 27, 30, and 33; and

(b) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 38, 41, and 44, said LCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 39, 42, and 45, said LCDR3 having the amino acid sequence
5 selected from any one of SEQ ID Nos: 40, 43, and 46.

In particular, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises (a) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively, and (b) LCDR1, LCDR2, and LCDR3 sequences of SEQ ID
10 NOs: 38, 39, and 40, respectively.

Other domains of the disclosure specifically binding TNF α include amino acids that have been mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in the CDR regions with the CDR regions depicted in the sequences described in TABLE 1. Suitably, other domains of the disclosure specifically binding TNF α include
15 mutant amino acid sequences wherein no more than 1, 2, 3, 4, 5 or 10 amino acids have been mutated by amino acid deletion, insertion or substitution in the CDR regions when compared with the CDR regions depicted in the sequences described in TABLE 1. Mutations, e.g., substitutions, may potentially be made at any residue within the set of CDRs, and may be within CDR1, CDR2 and/or CDR3.

20 Suitably, a second domain specifically binding TNF α of the multispecific antibody of the present disclosure comprises a heavy chain variable region (VH) and a light chain variable region (VL), wherein:

(a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence having
25 at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 25, 28, and 31; said HCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 26, 29, and 32; said HCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one
30 of SEQ ID NOs: 27, 30, and 33; and/or

(b) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 38, 41, and 44; said LCDR2 having the amino acid sequence having
5 at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 39, 42, and 45; said LCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 40, 43, and 46.

Suitably, said second domain specifically binding TNF α comprises: HCDR1, HCDR2,
10 and HCDR3 having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 25, 26, and 27, respectively, and/or LCDR1, LCDR2, and LCDR3 having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 38, 39, and 40, respectively.

In a further embodiment, said second domain specifically binding TNF α comprises a
15 heavy chain variable region VHB and a light chain variable region VLB.

Suitably, the present disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises a heavy chain variable region VHB and wherein said VHB is VH1A, VH1B, VH3 or VH4. In one embodiment, said second domain specifically binding
20 TNF α of the present disclosure comprises a heavy chain variable region VHB, wherein said VHB is VH4. In a preferred embodiment, said second domain specifically binding TNF α of the present disclosure comprises a heavy chain variable region VHB, wherein said VHB is VH3.

Suitably, the present disclosure provides the multispecific antibody comprising a first
25 domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises a light chain variable region VLB and wherein said VLB comprises V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 frameworks FR1 to FR3, and a framework FR4, which is selected from a V κ FR4, particularly V κ 1 FR4, V κ 3 FR4, and a V λ FR4. Suitable V λ FR4 are as set forth in SEQ
30 ID NO: 97 to SEQ ID NO: 103. In one embodiment, said second domain specifically binding

TNF α comprises V λ FR4 comprising the amino acid sequence having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 97 to SEQ ID NO: 103, preferably to SEQ ID NO: 97 or SEQ ID NO: 98, more preferably to SEQ ID NO: 97. Suitably, said second domain specifically binding TNF α comprises V λ FR4 comprising the amino acid sequence selected from any of SEQ ID NO: 97 to SEQ ID NO: 103, preferably V λ FR4 as set forth in SEQ ID NO: 97 or 98, more preferably V λ FR4 as set forth in SEQ ID NO: 98.

Thus, in one embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises:

- (a) the HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively, and the LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively;
- (b) a VHB, wherein said VHB is VH3 or VH4, preferably VH3; and
- (c) a VLB comprising a VL framework comprising V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 FR1 to FR3, and a framework FR4, which is selected from a V κ FR4, particularly V κ 1 FR4, V κ 3 FR4, and a V λ FR4, particularly V λ FR4 comprising the amino acid sequence having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 97 to SEQ ID NO: 103, preferably V λ FR4 as set forth in any of SEQ ID NO: 97 to SEQ ID NO: 103, preferably V λ FR4 as set forth in SEQ ID NO: 97 or 98, more preferably V λ FR4 as set forth in SEQ ID NO: 98.

Suitably, the present disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises a VH listed in TABLE 1. Suitably, the present disclosure also provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises (or alternatively, consisting of) a VH amino acid sequence listed in TABLE 1, wherein no more than about 20 amino acids, preferably no more than about 10 amino acids in a framework sequence (for example, a sequence which is not a CDR) have been mutated (wherein a mutation is, as various non-limiting examples, an addition, substitution or deletion). Other

domains of the disclosure specifically binding TNF α include amino acids that have been mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in the VH regions with the VH regions depicted in the sequences described in TABLE 1.

Suitably, the present disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises a VL domain listed in TABLE 1. Suitably, the present disclosure also provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises (or alternatively, consisting of) a VL amino acid sequence listed in TABLE 1, wherein no more than about 20 amino acids, preferably no more than about 10 amino acids in a framework sequence (for example, a sequence which is not a CDR) have been mutated (wherein a mutation is, as various non-limiting examples, an addition, substitution or deletion). Other domains of the disclosure specifically binding to TNF α include amino acids that have been mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in the VL regions with the VL regions depicted in the sequences described in TABLE 1.

In one embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence selected from the group consisting of SEQ ID NOs: 34, 35, 36 and 37, preferably SEQ ID NO: 34, and in particular wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively. In a preferred embodiment, said second domain specifically binding TNF α comprises a heavy chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 34, and wherein said heavy chain variable region comprises G56A, R82L, S85A, K86Q (AHO numbering), and in particular wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively. Suitably, said second domain specifically binding TNF α may comprise a heavy chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 36, and wherein said heavy chain variable region comprises A24K, G56A, R82L (AHO numbering), and in

particular wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively. Suitably, said second domain specifically binding TNF α may comprise a heavy chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 37, and wherein said heavy chain variable region comprises G56A, R82L, S85A, K86Q (AHO numbering), and in particular wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively.

In another embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence selected from the group consisting of SEQ ID NOs: 47, 48, 49, 50 and 51, preferably SEQ ID NO: 47, and in particular wherein said domain comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively.

In a preferred embodiment, said second domain specifically binding TNF α comprises a light chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 47, and wherein said light chain variable region comprises T22N, A51R, F89Y (AHO numbering), and in particular wherein said domain comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively. Suitably, said second domain specifically binding TNF α may comprise a light chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 49, and wherein said light chain variable region comprises T22N, D88E, S95G, E99A (AHO numbering), and in particular wherein said domain LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively. Suitably, said second domain specifically binding TNF α may comprise a light chain variable region comprising an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 50, and wherein said light chain variable region comprises T22N, A51R, F89Y, S95G, G141T, L145V (AHO numbering), and in particular wherein said domain LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively. Suitably, said second domain specifically binding TNF α may comprise a light chain variable region comprising an amino acid sequence that is at least 90 percent

identical to the amino acid sequence SEQ ID NO: 51, and wherein said light chain variable region comprises T22N, K50Q, A51R, F89Y, G141T, L145V (AHO numbering), and in particular wherein said domain LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively.

5 In a further embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 34; and a
10 light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 47.

Preferably, said second domain specifically binding TNF α comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93,
15 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 34; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 47, and wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27,
20 respectively, and/or LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively, preferably wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively.

Suitably, said second domain specifically binding TNF α comprises a heavy chain
25 variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 37; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 50 or 51, and wherein
30 said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively, and/or LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39,

and 40, respectively, preferably wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively.

5 In a specific embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises a VH comprising an amino acid sequence selected from the group consisting of SEQ ID NOs: 34, 35, 36 and 37; and/or a VL thereof comprising an amino acid sequence selected from the group consisting of SEQ ID NOs: 47, 48, 49, 50 and 51. In particular embodiment, the second domain specifically binding TNF α comprises a
10 VH sequence of SEQ ID NO: 34 and a VL sequence of SEQ ID NO: 47. In yet another particular embodiment, the second domain specifically binding TNF α comprises a VH sequence of SEQ ID NO: 37 and a VL sequence of SEQ ID NO: 50 or SEQ ID NO: 51.

In one embodiment, a domain that specifically binds to human TNF α is a domain that is described in TABLE 1. Other domains of the disclosure having a binding specificity for
15 human TNF α include those wherein the amino acids or nucleic acids encoding the amino acids have been mutated, yet have at least 60, 70, 80, 90 or 95 percent identity to the sequences described in TABLE 1. In one embodiment, it includes mutant amino acid sequences wherein no more than 1, 2, 3, 4 or 5 amino acids have been mutated in the variable regions when compared with the variable regions depicted in the sequence described in
20 TABLE 1, while retaining substantially the same activity.

In yet another embodiment, the present disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain comprises amino acid sequences that are homologous to the sequences described in TABLE 1, and said domain binds to human TNF α ,
25 and retains the desired functional properties of those domains described in TABLE 1.

In one embodiment, a domain of the disclosure specifically binding TNF α has a heavy chain variable region comprising HCDR1, HCDR2, and HCDR3 sequences and a light chain variable region comprising LCDR1, LCDR2, and LCDR3 sequences, wherein one or more of these CDR sequences have specified amino acid sequences based on the domains described

herein or conservative modifications thereof, and wherein the domains retain the desired functional properties of the antibodies of the disclosure.

Accordingly, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein
5 said second domain comprises (or consists of):

a heavy chain variable region (VH) comprising, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, wherein said HCDR1 is amino acid sequence SEQ ID No: 25, or a conservative variant thereof; said HCDR2 is amino acid sequence SEQ ID No: 26, or a conservative variant thereof; said HCDR3 is amino acid
10 sequence selected from any one of SEQ ID No: 27, or a conservative variant thereof; and

a light chain variable region (VL) comprising, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, wherein said LCDR1 is amino acid sequence SEQ ID No: 38, or a conservative variant thereof; said LCDR2 is amino acid sequence SEQ ID No: 39, or a conservative variant thereof; said LCDR3 having the amino
15 acid sequence SEQ ID No: 40, or a conservative variant thereof;

wherein said domain specifically binds to human TNF α and/or neutralize TNF α .

Exemplary domains specifically binding to human serum albumin (HSA)

The multispecific antibody of the disclosure comprises a third domain specifically
20 binding human serum albumin, wherein said domain comprises a heavy chain variable region (VH) and a light chain variable region (VL), wherein: (a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, and (b) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3.

25 Suitably, the multispecific antibody of the present disclosure may comprise a third binding domain having a third specificity different from the specificity of the first and second domains. Suitably, the multispecific antibody of the present disclosure may comprise a third domain specifically binding to human serum albumin (HSA). Thus, in one embodiment, the

multispecific antibody of the disclosure comprises a first domain specifically binding IL-17A, a second domain specifically binding TNF α and a third domain specifically binding to human serum albumin.

Suitable domains specifically binding human serum albumin for use in the multispecific antibody of the present disclosure include, but are not limited to, the humanized monoclonal antibodies or binding domains thereof whose sequences are listed in TABLE 1.

In one embodiment, the domain specifically binding to human serum albumin comprises a set of CDRs: HCDR1, HCDR2, HCDR3, LCDR1, LCDR2 and LCDR3, wherein the set of CDRs has 10 or fewer amino acid substitutions, e.g., 9 or fewer amino acid substitutions, 8 or fewer amino acid substitutions, 7 or fewer amino acid substitutions, 6 or fewer amino acid substitutions, 5 or fewer amino acid substitutions, 4 or fewer amino acid substitutions, 3 or fewer amino acid substitutions, 2 or fewer amino acid substitutions, 1 or 0 amino acid substitutions, preferably 0 amino acid substitutions, from a set of CDRs in which

- (a) HCDR1' is as set forth in SEQ ID NO: 52; HCDR2' is as set forth in SEQ ID NO: 53; HCDR3' is as set forth in SEQ ID NO: 54; LCDR1' is as set forth in SEQ ID NO: 62; LCDR2' is as set forth in SEQ ID NO: 63; LCDR3' is as set forth in SEQ ID NO: 64; or
- (b) HCDR1' is as set forth in SEQ ID NO: 73; HCDR2' is as set forth in SEQ ID NO: 74; HCDR3' is as set forth in SEQ ID NO: 75; LCDR1' is as set forth in SEQ ID NO: 83; LCDR2' is as set forth in SEQ ID NO: 84; LCDR3' is as set forth in SEQ ID NO: 85.

In particular, the domain specifically binding to human serum albumin comprises a VH CDR having an amino acid sequence of any one of the VH CDRs listed in TABLE 1. In particular, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said domain comprising (or alternatively, consisting of) one, two, three, or more VH CDRs having an amino acid sequence of any of the VH CDRs listed in TABLE 1.

Suitably, the domain specifically binding to human serum albumin comprises a heavy chain variable region (VH), wherein said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, wherein

- 5 (a) said HCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 52, 55, and 58; said HCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 53, 56, and 59; said HCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 54, 57, and 60; or
- 10 (b) said HCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 73, 76, and 79; said HCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 74, 77, and 80; said HCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 75, 78, and 81.

15 In a preferred embodiment, the domain specifically binding to human serum albumin comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 52, 53, and 54, respectively. In another embodiment, the domain specifically binding to human serum albumin comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 73, 74, and 75, respectively.

Suitably, the domain specifically binding to human serum albumin comprises a VL CDR having an amino acid sequence of any one of the VL CDRs listed in TABLE 1. In particular, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said domain comprises (or alternatively, consisting of) one, two, three or more VL CDRs having an amino acid sequence of any of the VL CDRs listed in TABLE 1.

Suitably, the domain specifically binding to human serum albumin comprises a light chain variable region (VL), wherein said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, wherein

- 25 (a) said LCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 62, 65, and 68; said LCDR2 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 63, 66, and 69; and said LCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 64, 67, and 70; or
- 30 (b) said LCDR1 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 83, 86, and 89; said LCDR2 is as set forth in the amino acid sequence selected

from any one of SEQ ID Nos: 84, 87, and 90; and said LCDR3 is as set forth in the amino acid sequence selected from any one of SEQ ID Nos: 85, 88, and 91.

In a preferred embodiment, the domain specifically binding to human serum albumin comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 62, 63, and 64,

5 respectively. In another embodiment, the domain specifically binding to human serum albumin comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 83, 84, and 85, respectively.

Suitably, the present disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a heavy chain
10 variable region (VH) and a light chain variable region (VL), wherein:

(a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 52, 55, and 58, said HCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 53, 56, and 59, said HCDR3 having the amino acid
15 sequence selected from any one of SEQ ID Nos: 54, 57, and 60; and said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 62, 65, and 68, said LCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 63, 66, and 69, said LCDR3 having the amino acid sequence selected from any one of SEQ ID
20 Nos: 64, 67, and 70; or

(b) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 73, 76, and 79, said HCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 74, 77, and 80, said HCDR3 having the amino acid
25 sequence selected from any one of SEQ ID Nos: 75, 78, and 81; and said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence selected from any one of SEQ ID Nos: 83, 86, and 89, said LCDR2 having the amino acid sequence selected from any one of SEQ ID Nos: 84, 87, and 90, said LCDR3 having the amino acid sequence selected from any one of SEQ ID
30 Nos: 85, 88, and 91.

In a preferred embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises (a) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 52, 53, and 54, respectively, and (b) LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 62, 63, and 64, respectively. In
 5 another embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises (a) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 73, 74, and 75, respectively, and (b) LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 83, 84, and 85, respectively.

Other domains of the disclosure specifically binding HSA include amino acids that have
 10 been mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in the CDR regions with the CDR regions depicted in the sequences described in TABLE 1. Suitably, other domains of the disclosure specifically binding HSA include mutant amino acid sequences wherein no more than 1, 2, 3, 4, 5 or 10 amino acids have been mutated by amino acid deletion, insertion or substitution in the CDR regions when compared with the
 15 CDR regions depicted in the sequences described in TABLE 1. Mutations, e.g., substitutions, may potentially be made at any residue within the set of CDRs, and may be within CDR1, CDR2 and/or CDR3.

Suitably, a third domain specifically binding HSA of the multispecific antibody of the present disclosure comprises: a heavy chain variable region (VH) and a light chain variable
 20 region (VL), wherein:

- (a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 52, 55, and 58; said HCDR2 having the amino acid sequence having at least 60, 70, 80, 90,
 25 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 53, 56, and 59; said HCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 54, 57, and 60; and/or
- (b) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence having at least
 30 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 62, 65, and 68; said LCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91,

92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 63, 66, and 69; said LCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 64, 67, and 70.

Suitably, the multispecific antibody comprises a third domain specifically binding HSA, wherein said third domain comprises: HCDR1, HCDR2, and HCDR3 having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID Nos: 52, 53, and 54, respectively, and/or LCDR1, LCDR2, and LCDR3 having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 62, 63, and 64, respectively.

Suitably, a third domain specifically binding HSA of the multispecific antibody of the present disclosure comprises: a heavy chain variable region (VH) and a light chain variable region (VL), wherein:

(a) said VH comprises, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, said HCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 73, 76, and 79; said HCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 74, 77, and 80; said HCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 75, 78, and 81; and/or

(b) said VL comprises, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, said LCDR1 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 83, 86, and 89; said LCDR2 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 84, 87, and 90; said LCDR3 having the amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to any one of SEQ ID NOs: 85, 88, and 91.

Suitably, the multispecific antibody comprises a third domain specifically binding HSA, wherein said third domain comprises: HCDR1, HCDR2, and HCDR3 having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID Nos: 73, 74, and 75, respectively, and/or LCDR1, LCDR2, and LCDR3 having at least 60, 70, 80, 90,

91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity to sequences of SEQ ID NOs: 83, 84, and 85, respectively.

In a further embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a heavy chain
5 variable region VHC and a light chain variable region VLC.

Suitably, the present disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a heavy chain variable region VHC and wherein said VHC is VH1A, VH1B, VH3 or VH4. In one embodiment, a third domain of the present disclosure specifically binding HSA comprises a
10 heavy chain variable region VHC, wherein said VHC is VH4. In a preferred embodiment, a third domain of the present disclosure specifically binding HSA comprises a heavy chain variable region VHC, wherein said VHC is VH3.

Suitably, the present disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a light chain variable
15 region VLC and wherein said VLC comprises V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 frameworks FR1 to FR3, and a framework FR4, which is selected from a V κ FR4, particularly V κ 1 FR4, V κ 3 FR4, and a V λ FR4. Suitable V λ FR4 are as set forth in SEQ ID NO: 97 to SEQ ID NO: 103. In one embodiment, said third domain comprises V λ FR4 comprising the amino acid sequence
20 having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 97 to SEQ ID NO: 103, preferably to SEQ ID NO: 97 or SEQ ID NO: 98, more preferably to SEQ ID NO: 97. Suitably, said third domain comprises V λ FR4 comprising the amino acid sequence selected from any of SEQ ID NO: 97 to SEQ ID NO: 103, preferably V λ FR4 as set forth in SEQ ID NO: 97 or 98, more preferably V λ FR4 as set forth in SEQ ID
25 NO: 98.

Thus, in a preferred embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises:

(a) the HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 52, 53, and 54, respectively, and the LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs:
30 62, 63, and 64, respectively;

- (b) a VHC, wherein said VHC is VH3 or VH4, preferably VH3; and
- (c) a VLC comprising a VL framework comprising V_{κ} frameworks FR1, FR2 and FR3, particularly $V_{\kappa 1}$ or $V_{\kappa 3}$ FR1 to FR3, preferably $V_{\kappa 1}$ FR1 to FR3, and a framework FR4, which is selected from a V_{κ} FR4, particularly $V_{\kappa 1}$ FR4, $V_{\kappa 3}$ FR4, and a V_{λ} FR4, particularly V_{λ} FR4 comprising the amino acid sequence having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 97 to SEQ ID NO: 103, preferably V_{λ} FR4 as set forth in amino acid sequence selected from any one of SEQ ID NO: 97 to SEQ ID NO: 103, preferably V_{λ} FR4 as set forth in SEQ ID NO: 97 or 98, more preferably V_{λ} FR4 as set forth in SEQ ID NO: 98.

In a further embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises:

- (i) the HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 73, 74, and 75, respectively, and the LCDR1, LCDR2, and LCDR3 sequences of SEQ ID Nos: 83, 84, and 85, respectively;
- (ii) a VHC, wherein said VHC is VH3 or VH4, preferably VH3; and
- (iii) a VLC comprising a VL framework comprising V_{κ} frameworks FR1, FR2 and FR3, particularly $V_{\kappa 1}$ or $V_{\kappa 3}$ FR1 to FR3, preferably $V_{\kappa 1}$ FR1 to FR3, and a framework FR4, which is selected from a V_{κ} FR4, particularly $V_{\kappa 1}$ FR4, $V_{\kappa 3}$ FR4, and a V_{λ} FR4, particularly V_{λ} FR4 comprising the amino acid sequence having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 97 to SEQ ID NO: 103, preferably V_{λ} FR4 as set forth in amino acid sequence selected from any one of SEQ ID NO: 97 to SEQ ID NO: 103, preferably V_{λ} FR4 as set forth in SEQ ID NO: 97 or 98, more preferably V_{λ} FR4 as set forth in SEQ ID NO: 98.

Suitably, the present disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a VH listed in TABLE 1. Suitably, the present disclosure also provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises (or alternatively, consisting of) a VH amino acid sequence listed in TABLE 1, wherein no more than about 20 amino acids, preferably no more than about 10 amino acids in a framework

sequence (for example, a sequence which is not a CDR) have been mutated (wherein a mutation is, as various non-limiting examples, an addition, substitution or deletion). Other domains of the disclosure specifically binding HSA include amino acids that have been mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in
5 the VH regions with the VH regions depicted in the sequences described in TABLE 1.

Suitably, the present disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a VL domain listed in TABLE 1. Suitably, the present disclosure also provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises (or
10 alternatively, consisting of) a VL amino acid sequence listed in TABLE 1, wherein no more than about 20 amino acids, preferably no more than about 10 amino acids in a framework sequence (for example, a sequence which is not a CDR) have been mutated (wherein a mutation is, as various non-limiting examples, an addition, substitution or deletion). Other domains of the disclosure specifically binding to HSA include amino acids that have been
15 mutated, yet have at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity in the VL regions with the VL regions depicted in the sequences described in TABLE 1.

In one embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93,
20 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 61, and in particular wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 52, 53, and 54, respectively.

In another embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a light chain
25 variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NOs: 71, and in particular wherein said domain comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 62, 63, and 64, respectively.

In a further embodiment, the disclosure provides the multispecific antibody comprising
30 a third domain specifically binding HSA, wherein said third domain comprises a heavy chain

variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 61; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 71.

In a preferred embodiment, the present disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 61; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 71, and wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 52, 53, and 54, respectively, and/or LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 62, 63, and 64, respectively, preferably wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 52, 53, and 54, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 62, 63, and 64, respectively.

In a specific embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a VH comprising an amino acid sequence SEQ ID NO: 61, and/or a VL thereof comprising an amino acid sequence SEQ ID NO: 71. In a preferred embodiment, the third domain specifically binding HSA comprises a VH sequence of SEQ ID NO: 61 and a VL sequence of SEQ ID NO: 71.

In yet another embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 82, and in particular wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 73, 74, and 75, respectively.

In another embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 92, and in particular wherein said domain comprises LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 83, 84, and 85, respectively.

In a further embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 82; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 92.

In a preferred embodiment, the present disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 82; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 92, and wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 73, 74, and 75, respectively, and/or LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 83, 84, and 85, respectively, preferably wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 73, 74, and 75, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 83, 84, and 85, respectively.

In a specific embodiment, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises a VH comprising an amino acid sequence SEQ ID NO: 82, and/or a VL thereof comprising an amino acid sequence SEQ ID NO: 92. In a preferred embodiment, the third domain specifically binding HSA comprises a VH sequence of SEQ ID NO: 82 and a VL sequence of SEQ ID NO: 92.

In one embodiment, a domain that specifically binds to human serum albumin is a domain that is described in TABLE 1. In one embodiment, a domain that specifically binds to HSA comprises an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence
5 selected from the group consisting of SEQ ID NOs: 72 and 93, preferably SEQ ID NO: 72. In one embodiment, a domain that specifically binds to HSA is as set forth in SEQ ID NO: 72 or SEQ ID NO: 93, preferably SEQ ID NO: 72.

Other domains of the disclosure having a binding specificity for HSA include those wherein the amino acids or nucleic acids encoding the amino acids have been mutated, yet
10 have at least 60, 70, 80, 90 or 95 percent identity to the sequences described in TABLE 1. In one embodiment, it includes mutant amino acid sequences wherein no more than 1, 2, 3, 4 or 5 amino acids have been mutated in the variable regions when compared with the variable regions depicted in the sequence described in TABLE 1, while retaining substantially the same activity.

15 In yet another embodiment, the present disclosure provides the multispecific antibody comprising a third domain specifically binding, wherein said third domain comprises amino acid sequences that are homologous to the sequences described in TABLE 1, and said domain binds to human serum albumin, and retains the desired functional properties of those domains described in TABLE 1.

20 In one embodiment, a domain of the disclosure specifically binding HSA has a heavy chain variable region comprising HCDR1, HCDR2, and HCDR3 sequences and a light chain variable region comprising LCDR1, LCDR2, and LCDR3 sequences, wherein one or more of these CDR sequences have specified amino acid sequences based on the domains described herein or conservative modifications thereof, and wherein the domains retain the desired
25 functional properties of the antibodies of the disclosure.

Accordingly, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises (or consists of):

a heavy chain variable region (VH) comprising, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, wherein said HCDR1 is amino acid
30 sequence SEQ ID No: 52, or a conservative variant thereof; said HCDR2 is amino acid

sequence SEQ ID No: 53, or a conservative variant thereof; said HCDR3 is amino acid sequence selected from any one of SEQ ID No: 54, or a conservative variant thereof; and

5 a light chain variable region (VL) comprising, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, wherein said LCDR1 is amino acid sequence SEQ ID No: 62, or a conservative variant thereof; said LCDR2 is amino acid sequence SEQ ID No: 63, or a conservative variant thereof; said LCDR3 having the amino acid sequence SEQ ID No: 64, or a conservative variant thereof;

wherein said domain specifically binds to human serum albumin.

10 The disclosure also provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain comprises (or consists of):

a heavy chain variable region (VH) comprising, in sequence, the three complementary determining regions HCDR1, HCDR2 and HCDR3, wherein said HCDR1 is amino acid sequence SEQ ID No: 73, or a conservative variant thereof; said HCDR2 is amino acid sequence SEQ ID No: 74, or a conservative variant thereof; said HCDR3 is amino acid sequence selected from any one of SEQ ID No: 75, or a conservative variant thereof; and

15 a light chain variable region (VL) comprising, in sequence, the three complementary determining regions LCDR1, LCDR2 and LCDR3, wherein said LCDR1 is amino acid sequence SEQ ID No: 83, or a conservative variant thereof; said LCDR2 is amino acid sequence SEQ ID No: 84, or a conservative variant thereof; said LCDR3 having the amino acid sequence SEQ ID No: 85, or a conservative variant thereof;

wherein said domain specifically binds to human serum albumin.

Further domains of the multispecific antibody of the present disclosure

25 Suitable domains for use in the multispecific antibody of the present disclosure in addition include, but are not limited to, the antibodies or binding domains thereof whose sequences are listed in TABLE 1 and which were further modified (modified versions thereof).

In one embodiment, the multispecific antibody comprises a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain is

optimized for expression in a mammalian cell, and has a heavy chain variable region and a light chain variable region, wherein one or more of these sequences have specified amino acid sequences based on the domains described herein or conservative modifications thereof, and wherein the domains retain the desired functional properties of the domains of the disclosure.

5 In one embodiment, the multispecific antibody comprises a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain is optimized for expression in a mammalian cell, and has a heavy chain variable region and a light chain variable region, wherein one or more of these sequences have specified amino acid sequences based on the domains described herein or conservative modifications thereof, and
10 wherein the domains retain the desired functional properties of the domains of the disclosure.

In one embodiment, the multispecific antibody comprises a third domain specifically binding HSA, wherein said third domain is optimized for expression in a mammalian cell and has a heavy chain variable region and a light chain variable region, wherein one or more of these sequences have specified amino acid sequences based on the domains described herein
15 or conservative modifications thereof, and wherein the domains retain the desired functional properties of the domains of the disclosure.

As used herein, the term, “optimized” means that a nucleotide sequence has been altered to encode an amino acid sequence using codons that are preferred in the production cell or organism, generally a eukaryotic cell, for example, a cell of Pichia, a Chinese Hamster Ovary
20 cell (CHO) or a human cell. The optimized nucleotide sequence is engineered to retain completely or as much as possible the amino acid sequence originally encoded by the starting nucleotide sequence, which is also known as the “parental” sequence. The optimized sequences herein have been engineered to have codons that are preferred in mammalian cells. However, optimized expression of these sequences in other eukaryotic cells or prokaryotic
25 cells is also envisioned herein. The amino acid sequences encoded by optimized nucleotide sequences are also referred to as optimized.

Accordingly, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain is optimized for expression in a mammalian cell, comprising a heavy chain
30 variable region and a light chain variable region wherein: the heavy chain variable region

comprises an amino acid sequence selected from any of SEQ ID NOs: 10 and 11, and conservative modifications thereof; and the light chain variable region comprises an amino acid sequence selected from any of SEQ ID NOs: 21 and 22, and conservative modifications thereof; wherein said first domain specifically binds to human IL-17A and/or neutralize IL-17A.

Accordingly, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said second domain is optimized for expression in a mammalian cell comprising a heavy chain variable region and a light chain variable region wherein: the heavy chain variable region comprises an amino acid sequence selected from any of SEQ ID NOs: 34, 35, 36 and 37, and conservative modifications thereof; and the light chain variable region comprises an amino acid sequence selected from any of SEQ ID NOs: 47, 48, 49, 50 and 51, and conservative modifications thereof; wherein said domain specifically binds to human TNF α and/or neutralize TNF α .

Accordingly, the disclosure provides the multispecific antibody comprising a third domain specifically binding HSA, wherein said third domain is optimized for expression in a mammalian cell comprising a heavy chain variable region and a light chain variable region wherein: the heavy chain variable region comprises an amino acid sequence selected from any of SEQ ID NO: 61, and SEQ ID NO: 82, and conservative modifications thereof; and the light chain variable region comprises an amino acid sequence selected from any of SEQ ID NO: 71, and SEQ ID NO: 92, and conservative modifications thereof; wherein said domain specifically binds to human serum albumin.

Another type of variable region modification is to mutate amino acid residues within the VH and/or VL CDR1, CDR2 and/or CDR3 regions to thereby improve one or more binding properties (e.g., affinity) of the antibody of interest, known as “affinity maturation”. Site-directed mutagenesis or PCR-mediated mutagenesis can be performed to introduce the mutation(s) and the effect on antibody binding, or other functional property of interest, can be evaluated in in vitro or in vivo assays as described herein and provided in the Examples. Conservative modifications (as discussed above) can be introduced. The mutations may be amino acid substitutions, additions or deletions. Moreover, typically no more than one, two, three, four or five residues within a CDR region are altered.

An “affinity-matured” antibody is one with one or more alterations in one or more variable domains thereof that result in an improvement in the affinity of the antibody for antigen, compared to a parent antibody that does not possess those alteration(s). In one embodiment, an affinity-matured antibody has nanomolar or even picomolar affinities for the target antigen. Affinity-matured antibodies are produced by procedures known in the art. For example, Marks et al, *Bio/Technology* 10:779-783 (1992) describes affinity maturation by VH- and VL-domain shuffling. Random mutagenesis of HVR and/or framework residues is described by, for example: Barbas et al. *Proc Natl. Acad. Sci. U.S.A* 91:3809-3813 (1994); Schier et al. *Gene* 169:147-155 (1995); Jackson et al, *J. Immunol.* 154(7):3310- 9 (1995); and Hawkins et al, *J. Mol. Biol.* 226:889-896 (1992). Accordingly, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A and a second domain specifically binding TNF α , wherein said first domain is an affinity-matured domain and/or said second domain is an affinity-matured domain. In a further embodiment, the disclosure provides the multispecific antibody further comprising a third domain specifically binding HSA, wherein said third domain is an affinity-matured domain.

An antibody of the disclosure further can be prepared using an antibody having one or more of the VH and/or VL sequences shown herein as starting material to engineer a modified antibody, which modified antibody may have altered properties from the starting antibody. An antibody can be engineered by modifying one or more residues within one or both variable regions (i.e., VH and/or VL), for example within one or more CDR regions and/or within one or more framework regions. Additionally or alternatively, an antibody can be engineered by modifying residues within the constant region(s), for example to alter the effector function(s) of the antibody.

One type of variable region engineering that can be performed is CDR grafting. Antibodies interact with target antigens predominantly through amino acid residues that are located in the six heavy and light chain complementarity determining regions (CDRs). For this reason, the amino acid sequences within CDRs are more diverse between individual antibodies than sequences outside of CDRs. Because CDR sequences are responsible for most antibody-antigen interactions, it is possible to express recombinant antibodies that mimic the properties of specific naturally occurring antibodies by constructing expression vectors that include CDR sequences from the specific naturally occurring antibody grafted onto framework sequences from a different antibody with different properties (see, e.g.,

Riechmann, L. et al., 1998 Nature 332:323-327; Jones, P. et al., 1986 Nature 321:522- 525; Queen, C. et al., 1989 Proc. Natl. Acad. Sci. U.S.A. 86: 10029-10033; U.S. Pat. No. 5,225,539 to Winter, and U.S. Pat. Nos. 5,530,101; 5,585,089; 5,693,762 and 6,180,370 to Queen et al.).

5 Such framework sequences can be obtained from public DNA databases or published references that include germline antibody gene sequences or rearranged antibody sequences. For example, germline DNA sequences for human heavy and light chain variable region genes can be found in the “VBase” human germline sequence database (available on the Internet at www.mrc-cpe.cam.ac.uk/vbase), as well as in Kabat, E. A., et al., 1991 Sequences
10 of Proteins of Immunological Interest, Fifth Edition, U.S. Department of Health and Human Services, NIH Publication No. 91-3242; Tomlinson, I. M., et al., 1992 J. Mol. Biol. 227:776-798; and Cox, J. P. L. et al., 1994 Eur. J Immunol. 24:827-836; the contents of each of which are expressly incorporated herein by reference. For example, germline DNA sequences for human heavy and light chain variable region genes and rearranged antibody sequences can be
15 found in “IMGT” database (available on the Internet at www.imgt.org; see Lefranc, M.P. et al., 1999 Nucleic Acids Res. 27:209-212; the contents of each of which are expressly incorporated herein by reference).

 An example of framework sequences for use in the binding domains of the multispecific antibodies of the disclosure are those that are structurally similar to the framework sequences
20 used by selected domains of the disclosure, e.g., consensus sequences and/or framework sequences used by domains of the disclosure. The VH CDR1, 2 and 3 sequences, and the VL CDR1, 2 and 3 sequences, can be grafted onto framework regions that have the identical sequence as that found in the germline immunoglobulin gene from which the framework sequence derive, or the CDR sequences can be grafted onto framework regions that contain
25 one or more mutations as compared to the germline sequences. For example, it has been found that in certain instances it is beneficial to mutate residues within the framework regions to maintain or enhance the antigen binding ability of the antibody (see e.g., U.S. Pat. Nos. 5,530,101; 5,585,089; 5,693,762 and 6,180,370 to Queen et al).

Exemplary multispecific antibodies of the present disclosure

In a specific embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A, a second domain specifically binding TNF α , and, optionally, a third domain specifically binding to HSA, wherein (i) said first domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively, and (ii) said second domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively, and (iii) optionally, said third domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 52, 53, and 54, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 62, 63, and 64, respectively.

In a more specific embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A, a second domain specifically binding TNF α , and, optionally, a third domain specifically binding to HSA, wherein

(i) said first domain specifically binding IL-17A of the multispecific antibody of the present disclosure comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 10; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 21, and wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively; and

(ii) said second domain specifically binding TNF α comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 34; and a light chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 47, and wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 25, 26, and 27, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively; and

(iii) optionally, said third domain comprises a heavy chain variable region comprising an amino acid sequence that is at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 61; and a light chain variable region comprising an amino acid sequence that is at least
5 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent, preferably at least 90 percent, identical to the amino acid sequence SEQ ID NO: 71, and wherein said domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 52, 53, and 54, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 62, 63, and 64, respectively.

In a specific embodiment, the disclosure provides the multispecific antibody comprising
10 a first domain specifically binding IL-17A, a second domain specifically binding TNF α and a third domain specifically binding to HSA, wherein said multispecific antibody comprises an amino acid sequence having at least 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98 or 99 percent identity, preferably at least 80% identity, more preferably at least 90% identity, to the sequence selected from any of SEQ ID NOs: 112, 113, 114, 115, 116, 117, 118, 119, 120,
15 121, 122, 123, and 124, preferably SEQ ID NO: 119, in particular wherein

(a) said first binding domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively;

(b) said second binding domain comprises HCDR1, HCDR2, and HCDR3
20 sequences of SEQ ID NOs: 25, 26, and 27, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 38, 39, and 40, respectively; and

(c) said third binding domain comprises HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 52, 53, and 54, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 62, 63, and 64, respectively.

25 In a more specific embodiment, the disclosure provides the multispecific antibody comprising a first domain specifically binding IL-17A, a second domain specifically binding TNF α and a third domain specifically binding to HSA, wherein said multispecific antibody comprises (or consists of) an amino acid sequence selected from any of SEQ ID NOs: 112, 113, 114, 115, 116, 117, 118, 119, 120, 121, 122, 123, and 124, preferably SEQ ID NO: 119.

30 ***Additional multispecific molecules of the disclosure***

The present disclosure also provides an antibody which is a multispecific molecule comprising an anti-IL-17A antibody of the present disclosure and at least another specificity, e.g., bispecific molecule, trispecific molecule, tetraspecific, pentaspecific, hexaspecific molecule.

5 The term “multispecific molecule” or “multispecific antibody” as used herein, refers to an antibody that binds to two or more different epitopes on at least two or more different targets (e.g., IL-17A and another target different from IL-17A), or binds to two or more different epitopes of the same target.

10 The term “multispecific molecule” includes bispecific, trispecific, tetraspecific, pentaspecific and hexaspecific antibodies. The term “bispecific antibody” as used herein, refers to an antibody that binds to two different epitopes on two different targets or on the same target. The term “trispecific antibody” as used herein, refers to an antibody that binds to three different epitopes on three different targets or on the same target.

15 An antibody of the disclosure can be derivatized or linked to another functional molecule, e.g., another peptide or protein (e.g., another antibody or ligand for a receptor) to generate a multispecific molecule that binds to at least two binding sites and/or different target molecules. The antibody of the disclosure may in fact be derivatized or linked to more than one other functional molecule to generate multispecific molecules that bind to more than two different binding sites and/or target molecules. To create a multispecific molecule of the
20 disclosure, an antibody of the disclosure can be functionally linked (e.g., by chemical coupling, genetic fusion, noncovalent association or otherwise) to one or more other binding molecules, such as another antibody, peptide or binding mimetic, such that a multispecific molecule results.

25 Accordingly, the present disclosure includes multispecific molecules comprising at least one first binding specificity for IL-17A and a second binding specificity for a second target epitope. For example, the second target epitope is present on another target molecule different from IL-17A. Accordingly, the present disclosure includes multispecific molecules comprising at least one first binding specificity for IL-17A and a second binding specificity for a second target epitope. For example, the second target epitope is another epitope of IL-

17A different from the first target epitope. The multispecific molecule can further include a third binding specificity, in addition to the first and second target epitope.

In a further embodiment, the present disclosure includes multispecific molecules monovalent, bivalent or multivalent for IL-17A specificity, preferably monovalent.

5 In another particular embodiment of the present disclosure, the antibody of the present disclosure is a monovalent or multivalent for IL-17A specificity molecule, e.g., bivalent, trivalent, tetravalent, pentavalent, hexavalent.

The term “monovalent molecule” or “monovalent antibody”, as used herein, refers to an antibody that has a single antigen-binding moiety that binds to a single epitope on a target
10 molecule, such as IL-17A.

The term “multivalent antibody” refers to a single binding molecule with more than one valency, where “valency” is described as the number of antigen-binding moieties that binds to epitopes on identical target molecules. As such, the single binding molecule can bind to more than one target molecule, or more than one binding site on a target molecule that contains
15 multiple copies of the epitope. Examples of multivalent antibodies include, but are not limited to bivalent antibodies, trivalent antibodies, tetravalent antibodies, pentavalent antibodies, and the like. The term “bivalent antibody” as used herein, refers to an antibody that has two antigen binding moieties, each of which binds to an identical epitope.

Suitably, the multispecific molecule of the present disclosure, e.g., bispecific molecule,
20 and / or a multivalent molecule, e.g., monovalent for IL-17A specificity molecule, bivalent for IL-17A specificity molecule, is an antibody format selected from any suitable multispecific, e.g. bispecific, format known in the art, including, by way of non-limiting example, formats based on a single-chain diabody (scDb), a tandem scDb (Tandab), a linear dimeric scDb (LD-scDb), a circular dimeric scDb (CD-scDb), a bispecific T-cell engager (BiTE; tandem di-scFv), a tandem tri-scFv, a tribody (Fab-(scFv)₂) or bibody (Fab-(scFv)₁), Fab, , Fab-Fv₂,
25 Morrison (IgG CH3-scFv fusion (Morrison L) or IgG CL-scFv fusion (Morrison H)), triabody, scDb-scFv, bispecific Fab₂, di-miniantibody, tetrabody, scFv-Fc-scFv fusion, scFv-HSA-scFv fusion, di-diabody, DVD-Ig, COVD, IgG-scFab, scFab-dsscFv, Fv₂-Fc, IgG-scFv fusions, such as bsAb (scFv linked to C-terminus of light chain), Bs1Ab (scFv linked to N-terminus of light chain), Bs2Ab (scFv linked to N-terminus of heavy chain), Bs3Ab (scFv
30

linked to C-terminus of heavy chain), Ts1Ab (scFv linked to N-terminus of both heavy chain and light chain), Ts2Ab (dsScFv linked to C-terminus of heavy chain), bispecific antibodies based on heterodimeric Fc domains, such as Knob-into-Hole antibodies (KiHs) (bispecific IgGs prepared by the KiH technology); an Fv, scFv, scDb, tandem-di-scFv, tandem tri-scFv, Fab-(scFv)₂, Fab-(scFv)₁, Fab, Fab-Fv₂, COVD fused to the N- and/or the C-terminus of either chain of a heterodimeric Fc domain or any other heterodimerization domain, a MATCH (described in WO 2016/0202457; Egan T., et al., mAbs 9 (2017) 68-84) and DuoBodies (bispecific IgGs prepared by the Duobody technology) (MAbs. 2017 Feb/Mar;9(2):182-212. doi: 10.1080/19420862.2016.1268307). Particularly suitable for use herein is a single-chain diabody (scDb) or scDb-scFv.

The term “diabodies” refers to antibody fragments with two antigen-binding sites, which fragments comprise a VH connected to VL in the same polypeptide chain (VH-VL). By using a linker that is too short to allow pairing between the two domains on the same chain, the domains are forced to pair with the complementary domains of another chain to create two antigen-binding sites. Diabodies may be bivalent or bispecific. Diabodies are described more fully in, for example, EP404097, WO 93/01161, Hudson et al., Nat. Med. 9:129-134 (2003), and Holliger et al., Proc. Natl. Acad. Sci. U.S.A. 90: 6444-6448 (1993). Triabodies and tetrabodies are also described in Hudson et al., Nat. Med. 9:129-134 (2003). The term “scDb-scFv” refers to an antibody format, wherein a single-chain Fv (scFv) fragment is fused by a flexible Gly-Ser linker to a single-chain diabody (scDb).

The bispecific scDb, in particular the bispecific monomeric scDb, particularly comprises two variable heavy chain domains (VH) or fragments thereof and two variable light chain domains (VL) or fragments thereof connected by linkers L1, L2 and L3 in the order VHA-L1-VLB-L2-VHB-L3-VLA, VHA-L1-VHB-L2-VLB-L3-VLA, VLA-L1-VLB-L2-VHB-L3-VHA, VLA-L1-VHB-L2-VLB-L3-VHA, VHB-L1-VLA-L2-VHA-L3-VLB, VHB-L1-VHA-L2-VLA-L3-VLB, VLB-L1-VLA-L2-VHA-L3-VHB or VLB-L1-VHA-L2-VLA-L3-VHB, wherein the VLA and VHA domains jointly form the antigen binding site for the first antigen, and VLB and VHB jointly form the antigen binding site for the second antigen.

The linker L1 particularly is a peptide of 2-10 amino acids, more particularly 3-7 amino acids, and most particularly 5 amino acids, and linker L3 particularly is a peptide of 1-10 amino acids, more particularly 2-7 amino acids, and most particularly 5 amino acids. The

middle linker L2 particularly is a peptide of 10-40 amino acids, more particularly 15-30 amino acids, and most particularly 20-25 amino acids.

In one embodiment, the multispecific and/or multivalent molecules of the present disclosure is in a MATCH format described in WO 2016/0202457; Egan T., et al., mAbs 9
5 (2017) 68-84.

Multispecific and/or multivalent molecules of the present disclosure can be produced using any convenient antibody manufacturing method known in the art (see, e.g., Fischer, N. & Leger, O., Pathobiology 74 (2007) 3-14 with regard to the production of bispecific constructs; Hornig, N. & Färber-Schwarz, A., Methods Mol. Biol. 907 (2012) 713-727, and
10 WO 99/57150 with regard to bispecific diabodies and tandem scFvs). Specific examples of suitable methods for the preparation of the bispecific construct of the present disclosure further include, inter alia, the Genmab (see Labrijn et al., Proc. Natl. Acad. Sci. U.S.A. 110 (2013) 5145-5150) and Merus (see de Kruif et al., Biotechnol. Bioeng. 106 (2010) 741-750) technologies. Methods for production of bispecific antibodies comprising a functional
15 antibody Fc part are also known in the art (see, e.g., Zhu et al., Cancer Lett. 86 (1994) 127-134); and Suresh et al., Methods Enzymol. 121 (1986) 210-228).

Other antibodies which can be employed in the multispecific and in the multivalent molecules of the disclosure are murine, chimeric and humanized monoclonal antibodies.

The multispecific molecules of the present disclosure can be prepared by conjugating the
20 constituent binding specificities, using methods known in the art. For example, each binding specificity of the bispecific molecule can be generated separately and then conjugated to one another. When the binding specificities are proteins or peptides, a variety of coupling or cross-linking agents can be used for covalent conjugation. Examples of cross-linking agents include protein A, carbodiimide, N-succinimidyl-5-acetyl-thioacetate (SATA), 5,5'-dithiobis
25 (2-nitrobenzoic acid) (DTNB), o-phenylenedimaleimide (oPDM), N-succinimidyl-3-(2-pyridyldithio)propionate (SPDP), and sulfosuccinimidyl 4-(N-maleimidomethyl)cyclohexane-1-carboxylate (sulfo-SMCC) (see e.g., Karpovsky et al., 1984 J. Exp. Med. 160: 1686; Liu, M A et al., 1985 Proc. Natl. Acad. Sci. U.S.A. 82:8648). Other methods include those described in Paulus, 1985 Behring Ins. Mitt. No. 78, 118-132; Brennan
30 et al., 1985 Science 229:81-83), and Glennie et al., 1987 J. Immunol. 139: 2367-2375).

Conjugating agents are SATA and sulfo-SMCC, both available from Pierce Chemical Co. (Rockford, 111.).

When the binding specificities are antibodies, they can be conjugated by sulfhydryl bonding of the C-terminus hinge regions of the two heavy chains. In a particularly
5 embodiment, the hinge region is modified to contain an odd number of sulfhydryl residues, for example one, prior to conjugation.

Alternatively, two or more binding specificities can be encoded in the same vector and expressed and assembled in the same host cell. This method is particularly useful where the bispecific molecule is a mAb X mAb, mAb X Fab, Fab X F (ab')₂ or ligand X Fab fusion
10 protein. Suitably, the multispecific molecule of the present disclosure can be a single chain molecule comprising one single chain antibody and a binding determinant, or a single chain multispecific molecule comprising two binding determinants. Multispecific molecules may comprise at least two single chain molecules. Methods for preparing multispecific molecules are described for example in U.S. Pat. No. 5,260,203; U.S. Pat. No. 5,455,030; U.S. Pat. No.
15 4,881,175; U.S. Pat. No. 5,132,405; U.S. Pat. No. 5,091,513; U.S. Pat. No. 5,476,786; U.S. Pat. No. 5,013,653; U.S. Pat. No. 5,258,498; and U.S. Pat. No. 5,482,858.

Binding of the multispecific molecules to their specific targets can be confirmed by, for example, enzyme-linked immunosorbent assay (ELISA), radioimmunoassay (REA), FACS analysis, bioassay (e.g., growth inhibition), or Western Blot assay. Each of these assays
20 generally detects the presence of protein-antibody complexes of particular interest by employing a labeled reagent (e.g., an antibody) specific for the complex of interest.

Nucleic acids, vectors, hosts cells and methods of production of the present disclosure

In a further aspect, the disclosure provides a nucleic acid encoding an antibody of the disclosure or a fragment or antigen-binding fragment thereof. The present disclosure also
25 provides nucleic acid sequences that encode CDRs, VH, VL, and antigen-binding fragments thereof that specifically bind to IL-17A protein. The present disclosure also provides nucleic acid sequences that encode CDRs, VH, VL, and antigen-binding fragments thereof that specifically bind to TNF α protein. The present disclosure also provides nucleic acid sequences that encode CDRs, VH, VL, and antigen-binding fragments thereof that

specifically bind to HSA protein. Such nucleic acid sequences can be optimized for expression in mammalian cells.

The term “nucleic acid” is used herein interchangeably with the term “polynucleotide” and refers to deoxyribonucleotides or ribonucleotides and polymers thereof in either single- or double-stranded form. The term encompasses nucleic acids containing known nucleotide analogs or modified backbone residues or linkages, which are synthetic, naturally occurring, and non-naturally occurring, which have similar binding properties as the reference nucleic acid, and which are metabolized in a manner similar to the reference nucleotides. Examples of such analogs include, without limitation, phosphorothioates, phosphoramidates, methyl phosphonates, chiral-methyl phosphorates, 2-O-methyl ribonucleotides, peptide-nucleic acids (PNAs). Unless otherwise indicated, a particular nucleic acid sequence also implicitly encompasses conservatively modified variants thereof (e.g., degenerate codon substitutions) and complementary sequences, as well as the sequence explicitly indicated. Specifically, as detailed below, degenerate codon substitutions may be achieved by generating sequences in which the third position of one or more selected (or all) codons is substituted with mixed-base and/or deoxyinosine residues (Batzner et al., *Nucleic Acid Res.* 19:5081, 1991; Ohtsuka et al., *J. Biol. Chem.* 260:2605-2608, 1985; and Rossolini et al., *Mol. Cell. Probes* 8:91-98, 1994).

The disclosure provides substantially purified nucleic acid molecules which encode polypeptides comprising segments or domains of the multispecific antibody chains described above and/or segments or domains of the IL-17A-binding antibody chains described above. When expressed from appropriate expression vectors, polypeptides encoded by these nucleic acid molecules are capable of exhibiting IL-17A and/or TNF α and/or HSA antigen binding capacity.

Also provided in the disclosure are polynucleotides which encode at least one CDR region and usually all three CDR regions from the heavy or light chain of the IL-17A-binding antibody set forth in TABLE 1, of the TNF α -binding domain set forth in TABLE 1, or of the HSA-binding domain set forth in TABLE 1. Some other polynucleotides encode all or substantially all of the variable region sequence of the heavy chain and/or the light chain of the IL-17A-binding antibody set forth in TABLE 1, of the TNF α -binding domain set forth in TABLE 1 or of the HSA-binding domain set forth in TABLE 1.. Because of the degeneracy

of the code, a variety of nucleic acid sequences will encode each of the immunoglobulin amino acid sequences.

The polynucleotide sequences can be produced by de novo solid-phase DNA synthesis or by PCR mutagenesis of an existing sequence (e.g., sequences as described in the Examples
5 below) encoding an IL-17A-binding antibody set forth in TABLE 1, a TNF α -binding domain set forth in TABLE 1 or an HSA-binding domain set forth in TABLE 1. Direct chemical synthesis of nucleic acids can be accomplished by methods known in the art, such as the phosphotriester method of Narang et al., 1979, Meth. Enzymol. 68:90; the phosphodiester method of Brown et al., Meth. Enzymol. 68: 109, 1979; the diethylphosphoramidite method
10 of Beaucage et al., Tetra. Lett., 22: 1859, 1981; and the solid support method of U.S. Pat. No. 4,458,066. Introducing mutations to a polynucleotide sequence by PCR can be performed as described in, e.g., PCR Technology: Principles and Applications for DNA Amplification, H. A. Erlich (Ed.), Freeman Press, NY, N.Y., 1992; PCR Protocols: A Guide to Methods and Applications, Innis et al. (Ed.), Academic Press, San Diego, Calif, 1990; Mattila et al.,
15 Nucleic Acids Res. 19:967, 1991; and Eckert et al., PCR Methods and Applications 1:17, 1991.

Also provided in the disclosure are expression vectors and host cells for producing the antibody of the disclosure.

The term “vector” is intended to refer to a polynucleotide molecule capable of
20 transporting another polynucleotide to which it has been linked. One type of vector is a “plasmid”, which refers to a circular double stranded DNA loop into which additional DNA segments may be ligated. Another type of vector is a viral vector, wherein additional DNA segments may be ligated into the viral genome. Certain vectors are capable of autonomous replication in a host cell into which they are introduced (e.g., bacterial vectors having a
25 bacterial origin of replication and episomal mammalian vectors). Other vectors (e.g., non-episomal mammalian vectors) can be integrated into the genome of a host cell upon introduction into the host cell, and thereby are replicated along with the host genome.

Moreover, certain vectors are capable of directing the expression of genes to which they are operatively linked. Such vectors are referred to herein as “recombinant expression
30 vectors” (or simply, “expression vectors”). In general, expression vectors of utility in

recombinant DNA techniques are often in the form of plasmids. In the present specification, “plasmid” and “vector” may be used interchangeably as the plasmid is the most commonly used form of vector. However, the disclosure is intended to include such other forms of expression vectors, such as viral vectors (e.g., replication defective retroviruses, adenoviruses and adeno-associated viruses), which serve equivalent functions.

The term “operably linked” refers to a functional relationship between two or more polynucleotide (e.g., DNA) segments. Typically, it refers to the functional relationship of a transcriptional regulatory sequence to a transcribed sequence. For example, a promoter or enhancer sequence is operably linked to a coding sequence if it stimulates or modulates the transcription of the coding sequence in an appropriate host cell or other expression system. Generally, promoter transcriptional regulatory sequences that are operably linked to a transcribed sequence are physically contiguous to the transcribed sequence, i.e., they are cis-acting. However, some transcriptional regulatory sequences, such as enhancers, need not be physically contiguous or located in close proximity to the coding sequences whose transcription they enhance.

Various expression vectors can be employed to express the polynucleotides encoding the antibody of the disclosure. Both viral-based and nonviral expression vectors can be used to produce the antibodies in a mammalian host cell. Nonviral vectors and systems include plasmids, episomal vectors, typically with an expression cassette for expressing a protein or RNA, and human artificial chromosomes (see, e.g., Harrington et al., *Nat Genet.* 15:345, 1997). For example, nonviral vectors useful for expression of the multispecific antibody of the disclosure or domains thereof, including of the IL-17A-binding polynucleotides and polypeptides, in mammalian (e.g., human) cells include pThioHis A, B and C, pcDNA3.1/His, pEBVHis A, B and C, (Invitrogen, San Diego, Calif.), MPS V vectors, and numerous other vectors known in the art for expressing other proteins. Useful viral vectors include vectors based on retroviruses, adenoviruses, adenoassociated viruses, herpes viruses, vectors based on SV40, papilloma virus, HBP Epstein Barr virus, vaccinia virus vectors and Semliki Forest virus (SFV). See, Brent et al., *supra*; Smith, *Annu. Rev. Microbiol.* 49:807, 1995; and Rosenfeld et al., *Cell* 68: 143, 1992.

The choice of expression vector depends on the intended host cells in which the vector is to be expressed. Typically, the expression vectors contain a promoter and other regulatory

sequences (e.g., enhancers) that are operably linked to the polynucleotides encoding the multispecific antibody of the disclosure or a domain thereof, including an IL-17A-binding antibody. In one embodiment, an inducible promoter is employed to prevent expression of inserted sequences except under inducing conditions. Inducible promoters include, e.g.,
5 arabinose, lacZ, metallothionein promoter or a heat shock promoter. Cultures of transformed organisms can be expanded under noninducing conditions without biasing the population for coding sequences whose expression products are better tolerated by the host cells. In addition to promoters, other regulatory elements may also be required or desired for efficient expression of the antibody of the disclosure or fragments thereof, including of an IL-17A-
10 binding antibody. These elements typically include an ATG initiation codon and adjacent ribosome binding site or other sequences. In addition, the efficiency of expression may be enhanced by the inclusion of enhancers appropriate to the cell system in use (see, e.g., Scharf et al., Results Probl. Cell Differ. 20: 125, 1994; and Bittner et al., Meth. Enzymol., 153:516, 1987). For example, the SV40 enhancer or CMV enhancer may be used to increase expression
15 in mammalian host cells.

The expression vectors may also provide a secretion signal sequence position to form a fusion protein with polypeptides encoded by inserted multispecific antibody-coding sequences, including IL-17A-binding antibody-coding sequences. More often, the inserted antibody sequences, e.g. multispecific antibody-coding or IL-17A-binding antibody-coding
20 sequences, are linked to signal sequences before inclusion in the vector. Vectors to be used to receive sequences encoding light and heavy chain variable domains of a multispecific antibody, or of fragments or domains thereof, including of an IL-17A-binding antibody, sometimes also encode constant regions or parts thereof. Such vectors allow expression of the variable regions as fusion proteins with the constant regions thereby leading to production of
25 intact antibodies. Typically, such constant regions are human.

The term "recombinant host cell" (or simply "host cell") refers to a cell into which a recombinant expression vector has been introduced. It should be understood that such terms are intended to refer not only to the particular subject cell but to the progeny of such a cell. Because certain modifications may occur in succeeding generations due to either mutation or
30 environmental influences, such progeny may not, in fact, be identical to the parent cell, but are still included within the scope of the term "host cell" as used herein.

The host cells for harboring and expressing the chains of the multispecific antibody, or of fragments or domains thereof, including of an IL-17A-binding antibody, can be either prokaryotic or eukaryotic. *E. coli* is one prokaryotic host useful for cloning and expressing the polynucleotides of the present disclosure. Other microbial hosts suitable for use include

5 bacilli, such as *Bacillus subtilis*, and other enterobacteriaceae, such as *Salmonella*, *Serratia*, and various *Pseudomonas* species. In these prokaryotic hosts, one can also make expression vectors, which typically contain expression control sequences compatible with the host cell (e.g., an origin of replication). In addition, any number of a variety of well-known promoters will be present, such as the lactose promoter system, a tryptophan (*trp*) promoter system, a

10 beta-lactamase promoter system, or a promoter system from phage lambda. The promoters typically control expression, optionally with an operator sequence, and have ribosome binding site sequences and the like, for initiating and completing transcription and translation. Other microbes, such as yeast, can also be employed to express the chains of the multispecific antibody, or of fragments or domains thereof, including of an IL-17A-binding antibody, of the

15 disclosure. Insect cells in combination with baculovirus vectors can also be used.

In one embodiment, mammalian host cells are used to express and produce the chains of the multispecific antibody, or of fragments or domains thereof, including of an IL-17A-binding antibody of the present disclosure. For example, they can be either a hybridoma cell line expressing endogenous immunoglobulin genes or a mammalian cell line harboring an

20 exogenous expression vector. These include any normal mortal or normal or abnormal immortal animal or human cell. For example, a number of suitable host cell lines capable of secreting intact immunoglobulins have been developed including the CHO cell lines, various Cos cell lines, HeLa cells, myeloma cell lines, transformed B-cells and hybridomas. The use of mammalian tissue cell culture to express polypeptides is discussed generally in, e.g.,

25 Winnacker, FROM GENES TO CLONES, VCH Publishers, N.Y., N.Y., 1987. Expression vectors for mammalian host cells can include expression control sequences, such as an origin of replication, a promoter, and an enhancer (see, e.g., Queen, et al., Immunol. Rev. 89:49-68, 1986), and necessary processing information sites, such as ribosome binding sites, RNA splice sites, polyadenylation sites, and transcriptional terminator sequences. These expression

30 vectors usually contain promoters derived from mammalian genes or from mammalian viruses. Suitable promoters may be constitutive, cell type-specific, stage-specific, and/or modulatable or regulatable. Useful promoters include, but are not limited to, the

metallothionein promoter, the constitutive adenovirus major late promoter, the dexamethasone-inducible MMTV promoter, the SV40 promoter, the MRP polIII promoter, the constitutive MPS V promoter, the tetracycline-inducible CMV promoter (such as the human immediate-early CMV promoter), the constitutive CMV promoter, and promoter-enhancer combinations known in the art.

Methods for introducing expression vectors containing the polynucleotide sequences of interest vary depending on the type of cellular host. For example, calcium chloride transfection is commonly utilized for prokaryotic cells, whereas calcium phosphate treatment or electroporation may be used for other cellular hosts. (See generally Sambrook, et al., supra). Other methods include, e.g., electroporation, calcium phosphate treatment, liposome-mediated transformation, injection and microinjection, ballistic methods, virosomes, immunoliposomes, polycation:nucleic acid conjugates, naked DNA, artificial virions, fusion to the herpes virus structural protein VP22 (Elliot and O'Hare, Cell 88:223, 1997), agent-enhanced uptake of DNA, and ex vivo transduction. For long-term, high-yield production of recombinant proteins, stable expression will often be desired. For example, cell lines which stably express polypeptides of the multispecific antibody, or of fragments or domains thereof, including of an IL-17A-binding antibody, can be prepared using expression vectors of the disclosure which contain viral origins of replication or endogenous expression elements and a selectable marker gene. Following the introduction of the vector, cells may be allowed to grow for 1-2 days in an enriched media before they are switched to selective media. The purpose of the selectable marker is to confer resistance to selection, and its presence allows growth of cells which successfully express the introduced sequences in selective media. Resistant, stably transfected cells can be proliferated using tissue culture techniques appropriate to the cell type. The present disclosure thus provides a method of producing an antibody of the disclosure, wherein said method comprises the step of culturing a host cell comprising a nucleic acid or a vector encoding the antibody of the disclosure, whereby said antibody of the disclosure or a fragment thereof is expressed.

Pharmaceutical compositions of the disclosure

In a further aspect, the present disclosure relates to a pharmaceutical composition comprising the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody

of the present disclosure, and a pharmaceutically acceptable carrier. Pharmaceutically acceptable carriers enhance or stabilize the composition, or facilitate preparation of the composition. Pharmaceutically acceptable carriers include solvents, dispersion media, coatings, antibacterial and antifungal agents, isotonic and absorption delaying agents, and the
5 like that are physiologically compatible.

A pharmaceutical composition of the present disclosure can be administered by a variety of methods known in the art. The route and/or mode of administration vary depending upon the desired results. Administration can be intravenous, intramuscular, intraperitoneal, or subcutaneous, or administered proximal to the site of the target. The pharmaceutically
10 acceptable carrier should be suitable for intravenous, intramuscular, subcutaneous, parenteral, spinal or epidermal administration (e.g., by injection or infusion). Depending on the route of administration, the active compound, i.e., antibody, and multispecific molecule, may be coated in a material to protect the compound from the action of acids and other natural conditions that may inactivate the compound.

15 Pharmaceutical compositions of the disclosure can be prepared in accordance with methods well known and routinely practiced in the art. See, e.g., Remington: The Science and Practice of Pharmacy, Mack Publishing Co., 20th ed., 2000; and Sustained and Controlled Release Drug Delivery Systems, J. R. Robinson, ed., Marcel Dekker, Inc., New York, 1978. Pharmaceutical compositions are preferably manufactured under GMP conditions. Typically,
20 a therapeutically effective dose or efficacious dose of the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, is employed in the pharmaceutical compositions of the disclosure. The multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific
25 molecule comprising an anti-IL-17A antibody of the present disclosure, is formulated into pharmaceutically acceptable dosage forms by conventional methods known to those of skill in the art. Dosage regimens are adjusted to provide the optimum desired response (e.g., a therapeutic response). For example, a single bolus may be administered, several divided doses may be administered over time or the dose may be proportionally reduced or increased as
30 indicated by the exigencies of the therapeutic situation. It is especially advantageous to formulate parenteral compositions in dosage unit form for ease of administration and uniformity of dosage. Dosage unit form as used herein refers to physically discrete units

suites as unitary dosages for the subjects to be treated; each unit contains a predetermined quantity of active compound calculated to produce the desired therapeutic effect in association with the required pharmaceutical carrier.

Actual dosage levels of the active ingredients in the pharmaceutical compositions of the present disclosure can be varied so as to obtain an amount of the active ingredient which is effective to achieve the desired therapeutic response for a particular patient, composition, and mode of administration, without being toxic to the patient. The selected dosage level depends upon a variety of pharmacokinetic factors including the activity of the particular compositions of the present disclosure employed, or the ester, salt or amide thereof, the route of administration, the time of administration, the rate of excretion of the particular compound being employed, the duration of the treatment, other drugs, compounds and/or materials used in combination with the particular compositions employed, the age, sex, weight, condition, general health and prior medical history of the patient being treated, and like factors.

Antibodies are usually administered on multiple occasions. Intervals between single dosages can be weekly, monthly or yearly. Intervals can also be irregular as indicated by measuring blood levels of the antibody in the patient. Alternatively, an antibody can be administered as a sustained release formulation, in which case less frequent administration is required. Dosage and frequency vary depending on the half-life of the antibody in the patient. In general, humanized antibodies show longer half-life than that of chimeric antibodies and nonhuman antibodies. The dosage and frequency of administration can vary depending on whether the treatment is prophylactic or therapeutic. In prophylactic applications, a relatively low dosage is administered at relatively infrequent intervals over a long period of time. Some patients continue to receive treatment for the rest of their lives. In therapeutic applications, a relatively high dosage at relatively short intervals is sometimes required until progression of the disease is reduced or terminated, and preferably until the patient shows partial or complete amelioration of symptoms of disease. Thereafter, the patient can be administered a prophylactic regime.

Uses and methods of the disclosure

The antibodies of the present disclosure have *in vitro* and *in vivo* diagnostic and therapeutic utilities. For example, these molecules can be administered to cells in culture, e.g.

in vitro or *in vivo*, or in a subject, e.g., *in vivo*, to treat, prevent or diagnose a variety of disorders.

The term “subject” includes human and non-human animals. Non-human animals include all vertebrates, e.g., mammals and non-mammals, such as non-human primates, sheep, dog,
5 cow, chickens, amphibians, and reptiles. Except when noted the terms “patient” or “subject” are used herein interchangeably.

The terms “treatment”, “treating”, “treat”, “treated”, and the like, as used herein, refer to obtaining a desired pharmacologic and/or physiologic effect. The effect may be therapeutic in terms of a partial or complete cure for a disease and/or adverse effect attributable to the
10 disease or delaying the disease progression. “Treatment”, as used herein, covers any treatment of a disease in a mammal, e.g., in a human, and includes: (a) inhibiting the disease, e.g., arresting its development; and (b) relieving the disease, e.g., causing regression of the disease.

The term “prevent” or “prevention” refers to a complete inhibition of development of a disease, or any secondary effects of disease. The term “prevent” or “prevention” as used
15 herein covers prevention of a disease or condition from occurring in an individual who may be predisposed to the disease but has not yet been diagnosed as having it.

The term “therapeutically effective amount” or “efficacious amount” refers to the amount of an agent that, when administered to a mammal or other subject for treating a disease, is sufficient to effect such treatment for the disease. The “therapeutically effective amount” will
20 vary depending on the agent, the disease and its severity and the age, weight, etc., of the subject to be treated.

In one aspect, the present disclosure relates to the antibody of the present disclosure, the multispecific molecule of the present disclosure, or the pharmaceutical composition of the present disclosure for use as a medicament.

25 In one aspect, the multispecific antibody of the present disclosure, or the pharmaceutical composition of the disclosure are particularly suitable for use in the treatment, prevention or diagnosis, in particular treatment, of a disorder mediated by IL-17A and/or TNF α or a disorder that can be treated by inhibiting Gro- α secretion.

In another aspect, the IL-17A-binding antibody of the disclosure or the pharmaceutical composition of the disclosure is particularly suitable for use in the treatment, prevention or diagnosis, in particular treatment, of a disorder mediated by IL-17A or a disorder that can be treated by inhibiting GRO- α secretion.

5 In the present disclosure, the term “a disorder mediated by IL-17A” encompasses all diseases and medical conditions in which IL-17A plays a role, whether directly or indirectly, in the disease or medical condition, including causation, development, progress, persistence or pathology of the disease or condition. Accordingly, the term “a disorder mediated by IL-17A” include conditions associated with or characterized by aberrant IL-17A levels and/or diseases
10 or conditions that can be treated by reducing or suppressing IL-17A induced activity in target cells or tissues, e.g., production of IL-6 or GRO- α . In the present disclosure, the term “a disorder mediated by TNF α ” encompasses all diseases and medical conditions in which TNF α plays a role, whether directly or indirectly, in the disease or medical condition, including causation, development, progress, persistence or pathology of the disease or condition.
15 Accordingly, the term “a disorder mediated by TNF α ” include conditions associated with or characterized by aberrant TNF α levels and/or diseases or conditions that can be treated by reducing or suppressing TNF α induced activity in target cells or tissues, e.g., production of Gro- α . Disorders mediated by IL-17A and/or TNF α include inflammatory conditions and autoimmune diseases, such as arthritis, rheumatoid arthritis, or psoriasis. These further
20 include allergies and allergic conditions, hypersensitive reactions, chronic obstructive pulmonary disease, cystic fibrosis and organ or tissue transplant rejection. For example, an antibody of the disclosure may be used for the treatment of recipients of heart, lung, combined heart-lung, liver, kidney, pancreatic, skin or corneal transplants, including allograft rejection or xenograft rejection, and for the prevention of graft-versus-host disease, such as following
25 bone marrow transplant, and organ transplant associated arteriosclerosis.

In one embodiment, the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure are particularly suitable for use in the treatment, prevention or diagnosis, in particular treatment,
30 of an inflammatory condition or an autoimmune disease.

The multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure is suitable for use in the treatment, prevention, or amelioration of autoimmune disease and of inflammatory conditions, in particular inflammatory conditions with an aetiology including an autoimmune component such as arthritis (for example rheumatoid arthritis, arthritis chronica progrediente and arthritis deformans) and rheumatic diseases, including inflammatory conditions and rheumatic diseases involving bone loss, inflammatory pain, spondyloarthropathies including ankylosing spondylitis, Reiter syndrome, reactive arthritis, psoriatic arthritis, juvenile idiopathic arthritis and enteropathic arthritis, enthesitis, hypersensitivity (including both airways hypersensitivity and dermal hypersensitivity) and allergies. Specific autoimmune diseases for which antibodies of the disclosure may be employed include autoimmune haematological disorders (including e.g. hemolytic anaemia, aplastic anaemia, pure red cell anaemia and idiopathic thrombocytopenia), systemic lupus erythematosus (SLE), lupus nephritis, inflammatory muscle diseases (dermatomyositis), periodontitis, polychondritis, scleroderma, Wegener granulomatosis, dermatomyositis, chronic active hepatitis, myasthenia gravis, psoriasis, Steven-Johnson syndrome, idiopathic sprue, autoimmune inflammatory bowel disease (including e.g. ulcerative colitis, Crohn's disease and irritable bowel syndrome), endocrine ophthalmopathy, Graves' disease, sarcoidosis, multiple sclerosis, systemic sclerosis, fibrotic diseases, primary biliary cirrhosis, juvenile diabetes (diabetes mellitus type I, type 1 diabetes), uveitis, keratoconjunctivitis sicca and vernal keratoconjunctivitis, interstitial lung fibrosis, periprosthetic osteolysis, glomerulonephritis (with and without nephrotic syndrome, e.g. including idiopathic nephrotic syndrome or minimal change nephropathy), multiple myeloma other types of tumors, inflammatory disease of skin and cornea, myositis, loosening of bone implants, metabolic disorders, (such as obesity, atherosclerosis and other cardiovascular diseases including dilated cardiomyopathy, myocarditis, diabetes mellitus type II, and dyslipidemia), and autoimmune thyroid diseases (including Hashimoto thyroiditis), small and medium vessel primary vasculitis, large vessel vasculitides including giant cell arteritis, hidradenitis suppurativa, neuromyelitis optica, Sjogren's syndrome, Behcet's disease, atopic and contact dermatitis, bronchiolitis, inflammatory muscle diseases, autoimmune peripheral neuropathies, immunological renal, hepatic and thyroid diseases, inflammation and atherothrombosis, autoinflammatory fever syndromes, immunohematological disorders, and bullous diseases of the skin and mucous

membranes. Anatomically, uveitis can be anterior, intermediate, posterior, or pan-uveitis. It can be chronic or acute. The etiology of uveitis can be autoimmune or non-infectious, infectious, associated with systemic disease, or a white-dot syndrome.

In a particular embodiment, the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure, is suitable for use in the treatment of multiple sclerosis, psoriasis, asthma, systemic lupus erythematosus (SLE), and lupus nephritis. In another specific embodiment, the antibody of the disclosure is suitable for use in the treatment of diabetes, in particular diabetes mellitus type I or type II.

The term “a disorder mediated by IL-17A and/or TNF α ” also includes inflammation-associated cancer. In a particular embodiment, the antibody of the disclosure is suitable for use in the treatment of cancer, in particular IL-17A- and/or TNF α -mediated cancer. In a particular embodiment, the antibody of the disclosure is suitable for use in the treatment of inflammation-associated cancer.

The term “cancer” refers to a disease characterized by the rapid and uncontrolled growth of aberrant cells. Cancer cells can spread locally or through the bloodstream and lymphatic system to other parts of the body. Examples of various cancers are described herein and include but are not limited to, breast cancer, prostate cancer, ovarian cancer, cervical cancer, skin cancer, pancreatic cancer, colorectal cancer, renal cancer, liver cancer, brain cancer, lymphoma, leukemia, lung cancer and the like. The terms “tumor” and “cancer” are used interchangeably herein, e.g., both terms encompass solid and liquid, e.g., diffuse or circulating, tumors. As used herein, the term “cancer” or “tumor” includes premalignant, as well as malignant cancers and tumors.

Non-limiting examples of inflammation-associated cancers include gastric cancers, colorectal cancers, non-small cell lung cancers, hepatocellular carcinomas and adenocarcinomas (Wu et al., 2014 Tumour Biol. 35(6):5347-56; Wu et al., 2012 PLoS One 7(12); Zhang et al. 2012 Asian Pac J Cancer Prev 13(8):3955-60; Liu et al., 2011 Biochem Biophys Res Commun. 407(2):348-54).

The multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, is also suitable for use in the diagnosis and/or prognosis of certain cancers, such as inflammation-associated cancers for example gastric cancers, colorectal
5 cancers, non-small cell lung cancers, hepatocellular carcinomas and adenocarcinomas. For example, IL-17A has been linked to the prognosis and poor survival in patients suffering from non-small cell lung cancer (Chen et al., 2010 Lung Cancer 69(3):348-54), colorectal carcinoma and hepatocellular carcinoma (Punt et al., 2015 Oncoimmunology, 4(2): e984547).

Thus, in a particular embodiment, the multispecific antibody of the present disclosure, an
10 anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, is suitable for use in the treatment of a cancer, arthritis, rheumatoid arthritis, osteoarthritis, reactive arthritis, psoriasis, chronic obstructive pulmonary disease, systemic lupus erythematosus (SLE), lupus nephritis, an autoimmune inflammatory bowel disease, asthma, multiple sclerosis, or cystic fibrosis, bone
15 loss, airways hypersensitivity, a demyelinating disorder, dermal hypersensitivity, acute transplant rejection, allograft rejection, graft-versus host disease, systemic sclerosis, an urological inflammatory disorder, a cardiovascular disease, vasculitis, a periodic fever, a glucose metabolism disorder, a pulmonary disease, periodontitis, hepatic stromal keratitis, an allergy, inflammatory pain, a spondyloarthropathy, septicaemia, septic or endotoxic shock,
20 meningitis, surgical trauma, an autoimmune haematological disorder, Alzheimer's disease, sarcoidosis, cirrhosis, hepatitis, glomerulonephritis or dislipidemia.

In one aspect, the present disclosure relates to use of the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical
25 composition of the disclosure, in a manufacture of a medicament.

In another aspect, the present disclosure relates to use of the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure, in a manufacture of a medicament for use in the treatment,
30 prevention or diagnosis, in particular treatment, of a disorder mediated by IL-17A and/or TNF α or a disorder that can be treated by inhibiting GRO- α secretion. In one embodiment,

the present disclosure relates to use of the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure, in a manufacture of a medicament for use in the treatment, prevention or
5 diagnosis, in particular treatment, of an inflammatory condition or an autoimmune disease. In another embodiment, the present disclosure relates to use of the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure, in a manufacture of a medicament for use in the treatment,
10 prevention or diagnosis, in particular treatment, of multiple sclerosis, psoriasis, asthma, systemic lupus erythematosus (SLE), and lupus nephritis. In a further embodiment, the present disclosure relates to use of the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the
15 disclosure, in a manufacture of a medicament for use in the treatment, prevention or diagnosis, in particular treatment, of diabetes, in particular diabetes mellitus type I or type II. In a further embodiment, the present disclosure relates to use of the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a
20 pharmaceutical composition of the disclosure, in a manufacture of a medicament for use in the treatment, prevention or diagnosis, in particular treatment, of cancer, in particular IL-17A and/or TNF α -mediated cancer, in a particular inflammation-associated cancer.

In another aspect, the present disclosure relates to a method for the treatment of a subject in need of therapy, characterized by administering to the subject a therapeutically effective
25 amount of multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure.

In a further aspect, the present disclosure relates to a method of treating a disorder mediated by IL-17A and/or TNF α , said method comprising administering an effective amount
30 of multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure, such that the condition

is alleviated. In one embodiment, the present disclosure relates to a method of treating an inflammatory condition or an autoimmune disease, said method comprising administering an effective amount of the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure, such that the condition is alleviated. In another embodiment, the present disclosure relates to a method of treating multiple sclerosis, psoriasis, asthma, systemic lupus erythematosus (SLE), and lupus nephritis, said method comprising administering an effective amount of the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure, such that the condition is alleviated. In further embodiment, the present disclosure relates to a method of treating diabetes, in particular diabetes mellitus type I or type II, said method comprising administering an effective amount of the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure, such that the condition is alleviated. In further embodiment, the present disclosure relates to a method of treating cancer, in particular IL-17A- and/or TNF α -mediated cancer, in particular inflammation-associated cancer, said method comprising administering an effective amount of the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure, such that the condition is alleviated.

Also within this disclosure is a kit comprising the multispecific antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, or a pharmaceutical composition of the disclosure. The kit can include one or more other elements including: instructions for use; other reagents, e.g., a label, a therapeutic agent, or an agent useful for chelating, or otherwise coupling, an antibody to a label or therapeutic agent, or a radioprotective composition; devices or other materials for preparing the antibody molecule for administration; pharmaceutically acceptable carriers; and devices or other materials for administration to a subject. In a specific embodiment, the kit comprises the multispecific

antibody of the present disclosure, an anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, in a pharmaceutically effective amount. In a further embodiment, the kit comprises a pharmaceutically effective amount of the multispecific antibody of the present disclosure, an
5 anti-IL-17A antibody of the present disclosure, or another multispecific molecule comprising an anti-IL-17A antibody of the present disclosure, in lyophilized form and a diluent and, optionally, instructions for use. Said kit may further comprise a filter needle for reconstitution and a needle for injecting.

TABLE 1 Sequence listing.

SEQ ID NO:	Description	Sequence
Anti-IL-17A Antibody 27-07-G02		
1	HCDR1 (H27-H42; AHo numbering)	GFSSSDYWMC
2	HCDR2 (H57-H76; AHo numbering)	CIYAGDVDDTDYASWARG
3	HCDR3 (H108-H138; AHo numbering)	RVDGFDITYFDL
4	HCDR1 (Kabat)	SDYWMC
5	HCDR2 (Kabat)	CIYAGDVDDTDYASWARG
6	HCDR3 (Kabat)	VDGFDITYFDL
7	HCDR1 (Chothia)	GFSSSDY
8	HCDR2 (Chothia)	AGDVD
9	HCDR3 (Chothia)	DGFDITYFD
10	VH (VH3; CDR-graft; from A1)	EVQLVESGGGLVQPGGSLRLSCAASGFSSSDYWMCWVVRQAPGKGLEWIGCIYAGDVDDT DYASWARGRFTISRDN SKNTVY LQMNSLRAEDTAVYYCARVDGFDITYFDLWGQGTLVTV SS
11	VH (VH3; STR-graft; from A2) Mutations (AHo): Q14K; G16E; G56A	EVQLVESGGGLVKPEGSLRLSCAASGFSSSDYWMCWVVRQAPGKGLEWIGCIYAGDVDDT DYASWARGRFTISRDN SKNTVY LQMNSLRAEDTAVYYCARVDGFDITYFDLWGQGTLVTV SS
12	LCDR1 (L24-L42; AHo numbering)	QASQTINYWLA
13	LCDR2 (L58-L72; AHo numbering)	RASTLAS
14	LCDR3 (L107-L138; AHo numbering)	QNYYYSTSRSYG

15	LCDR1 (Kabat)	QASQTINYWLA
16	LCDR2 (Kabat)	RASTLAS
17	LCDR3 (Kabat)	QNYYSSTRSYG
18	LCDR1 (Chothia)	SQTINYW
19	LCDR2 (Chothia)	RAS
20	LCDR3 (Chothia)	YYYSSTRSY
21	VL (Vk1-sk12; CDR-graft; from A1)	DIQMTQSPSSLSASVGDRVTITCQASQTINYWLA WYQQKPGKAPKLLIYRASTLASGVPS SGSGSGTDFTLTISSLQPEDFATYYCQNYYSSTRSYGF GGGTKLTVLG
22	VL (Vk1-sk12; STR-graft; from A2) Mutations (AHO): A51P	DIQMTQSPSSLSASVGDRVTITCQASQTINYWLA WYQQKPGKPPKLLIYRASTLASGVPS SGSGSGTDFTLTISSLQPEDFATYYCQNYYSSTRSYGF GGGTKLTVLG
23	Linker	GGGSGGGSGGGSGGGGS
24	A1 (scFv; VL-linker-VH)	DIQMTQSPSSLSASVGDRVTITCQASQTINYWLA WYQQKPGKAPKLLIYRASTLASGVPS SGSGSGTDFTLTISSLQPEDFATYYCQNYYSSTRSYGF GGGTKLTVLGGGGSGGGSGC GGSGGGSEVQLVESGGGLVQPGGSLRLSCAASGFS SDYMCWVRQAPGKGLEWIG YAGDVDDTDYASWARGRFTISRDNSKNTVYLMNS LRAEDTAVYYCARVDGFDITYFL WGQGTLLVTVSS
25	A2 (scFv; VL-linker-VH)	DIQMTQSPSSLSASVGDRVTITCQASQTINYWLA WYQQKPGKPPKLLIYRASTLASGVPS SGSGSGTDFTLTISSLQPEDFATYYCQNYYSSTRSYGF GGGTKLTVLGGGGSGGGSGC GGSGGGSEVQLVESGGGLVQPGGSLRLSCAASGFS SDYMCWVRQAPGKGLEWIA YAGDVDDTDYASWARGRFTISRDNSKNTVYLMNS LRAEDTAVYYCARVDGFDITYFL WGQGTLLVTVSS
Vλ germline-based FR4		
26	Vλ germline-based FR4, Sk17	FGTGTKVTVLG
27	Vλ germline-based FR4, Sk12	FGGGTKLTVLG
28	Vλ germline-based FR4	FGGGTQLIILG

29	Vλ germline-based FR4	FGEGTELTVLG
30	Vλ germline-based FR4	FGSGTKVTVLG
31	Vλ germline-based FR4	EGGGTQLTVLG
32	Vλ germline-based FR4	EGGGTQLTALG
IL-17 family		
33	IL-17A UniProt ID NO: Q16552	MTPGKTSLVSLLLLLSLEAIVKAGIAIPQNPQPCNSDKNFPRTVMVNLNIHNRNTNTNPK SDYYNRSTSPWNLRNEDPERYPSVIWEAKCRHLGC'VNADGKVDYHMHNSVPIQQEILVI REPPHCNPSFRLEKILVSVGCTCVTPIVHHVS
34	IL-17B UniProt ID NO: Q9UHF5	MDWPHNLLFLTISIFLGLGQPRSPKSKRGQGRPGPLAPGPHQVPLDLVSRMKPYARMII YERNIEEMVAQLRNSELQAQRKCEVNLQLWMSNKRSLSPWGYSHNDPSRIPVDLPEARC CLGCVNPFTMQEDRSMVSVPVFSQVVPVRRRLCPPPRTGPCRQRAVMETIAVGGCTCIF
35	IL-17C UniProt ID NO: Q9P0M4	MTLLPGLLFLTWLHTCLAHHDPSLRGHPHSHGTPHCYSAEELPLGQAPPHLLARGAKWC ALPVVALVSSLEAAASHRGRHERPSATTQCPVLRPEEVLEADTHQRSISPWRYRVDTDDEY QKLAFACELCRGCIDARTGRETAAALNSVRLQLSLLVLRRRPCSRDGSGLPTPGAFAFHTE HVPVGGCTCVLPRSV
36	IL-17D UniProt ID NO: Q8TAD2	MLVAGFLLALPPSWAAGAPRAGRPARPRGCADRPEELLEQLYGRLAAGVLSAFHHTLC GPREQARNASCPAGGRPADRRFRPPTNLRVSPWAYRISYDPARYPRYLPEAYCLCRGCI GLFGEEDVRFERSAPVYMPVTVLRRTACAGGRSVYTEAYVTIPVGGCTCVPEPEKDADSIN IDKQGAKLLLGPNDA PAGP
37	IL-17E UniProt ID NO: Q9H293	MREPRLGEDSSLISLFLQVVAFLAMVMGTHTYSHWPSCCPSKGGQDTSEELLRWSTVPV LEPARPNRHPESCRA SEDGPLNSRAISPWR YELDRDLNRLPQDL YHARCLCPHCVSLQTG MDPRGNSELL YHNQTVFYRRPCHGEKGTGKGYCLERRLYRVSACVCVRPRVMG
38	IL-17F UniProt ID NO: Q96PD4	MTVKTLHGPMVKYLLLSILGLAFLSEAAARKIPKVGHHTFFQKPESCIPPVPGGSMKLDIG ENQRVSMRSRNIESRSTSPWN'YTVTWDPNRYPSVVQAQCRNLGCINAAQKEDISMNSVP QETLVVRRKHQGCVSFQLEKVLTVVGCTCVTPVHHVQ
Anti-IL17A Antibody 27-31-C04		
39	HCDR1 (H27-H42; AHo numbering)	GFSGFGFYMC
40	HCDR2 (H57-H76; AHo numbering)	CIDTGDGTTYASWAKG

41	HCDR3 (H108-H138; AHo numbering)	RDAA YGGY AYP AHGMDL
42	HCDR1 (Kabat)	GFYYMC
43	HCDR2 (Kabat)	CIDTGDGTTYASWAKG
44	HCDR3 (Kabat)	DAAYGGY AYP AHGMDL
45	HCDR1 (Chothia)	GFSGFY
46	HCDR2 (Chothia)	TGDG
47	HCDR3 (Chothia)	AA YGGY AYP AHGMD
48	VH (VH3; CDR-graft; from PRO571)	EVQLVESGGGLVQPGGSLRLSCAASGFSFGFYMCWVRQAPGKGLEWIGCIDTGDGTT ASWAKGRFTISRDN SKNTVYLQMNSLRAEDTAVYYCARDAA YGGY AYP AHGMDLWG TLVTVSS
49	VH (VH3; STR-graft; from PRO592) Mutations (AHo): R20T; Q141P	EVQLVESGGGLVQPGGSLRLSCAASGFSFGFYMCWVRQAPGKGLEWIGCIDTGDGTT ASWAKGRFTISRDN SKNTVYLQMNSLRAEDTAVYYCARDAA YGGY AYP AHGMDLWG TLVTVSS
50	LCDR1 (L24-L42; AHo numbering)	QASEIYRYLS
51	LCDR2 (L58-L72; AHo numbering)	LASTLTS
52	LCDR3 (L107-L138; AHo numbering)	QSNFGTASTTYNT
53	LCDR1 (Kabat)	QASEIYRYLS
54	LCDR2 (Kabat)	LASTLTS
55	LCDR3 (Kabat)	QSNFGTASTTYNT
56	LCDR1 (Chothia)	SESIYRY
57	LCDR2 (Chothia)	LAS
58	LCDR3 (Chothia)	NFGTASTTYN

59	VL (Vk1-sk12; CDR-graft; from PRO571)	DIQMTQSPSSLSASVGDRVTITCQASESIYR YLSWYQQKPKAPKLLIYLA STLTSGVPSRI GSGGTDFTLTISSLQPEDFATYQCQSNFGTASTTYNTFTGGGKLTVLG
60	VL (Vk1-sk12; STR-graft; from PRO592)	DIQMTQSPSSLSASVGDRVTITCQASESIYR YLSWYQQKPKAPKLLIYLA STLTSGVPSRI GSGGTDFTLTISSLQPEDFATYQCQSNFGTASTTYNTFTGGGKLTVLG
61	PRO571 (scFv; VL-linker-VH)	DIQMTQSPSSLSASVGDRVTITCQASESIYR YLSWYQQKPKAPKLLIYLA STLTSGVPSRI GSGGTDFTLTISSLQPEDFATYQCQSNFGTASTTYNTFTGGGKLTVLG GGGGGGSEVQLVESGGGLVQPGGSLRLSCAASGFSFGFYMCWVRQAPGKLEWIG TGDGTTYASWAKGRFTISRDNKNTVYLMNSLR AEDTAVYYCARDAAAYGGYAYPAI MDLWGQGTLVTVSS
62	PRO592 (scFv; VL-linker-VH)	DIQMTQSPSSLSASVGDRVTITCQASESIYR YLSWYQQKPKAPKLLIYLA STLTSGVPSRI GSGGTDFTLTISSLQPEDFATYQCQSNFGTASTTYNTFTGGGKLTVLG GGGGGGSEVQLVESGGGLVQPGGSLRLSCAASGFSFGFYMCWVRQAPGKLEWIG TGDGTTYASWAKGRFTISRDNKNTVYLMNSLR AEDTAVYYCARDAAAYGGYAYPAI MDLWGQGTLVTVSS
Anti-TNFα domain (16-19-B11-sc06)		
63	HCDR1 (H27-H42; AHo numbering)	GIDFSTYGIS
64	HCDR2 (H57-H76; AHo numbering)	YIYPDYGITDYASWVNG
65	HCDR3 (H108-H138; AHo numbering)	RSGSYYSRGWGAHYFNL
66	HCDR1 (Kabat)	TYGIS
67	HCDR2 (Kabat)	YIYPDYGITDYASWVNG
68	HCDR3 (Kabat)	SGSYYSRGWGAHYFNL

69	HCDR1 (Chothia)		GIDFSTY
70	HCDR2 (Chothia)		PDYG
71	HCDR3 (Chothia)		GSYYSRGWGAHYFN
72	VH16-19-B11-sc06 (VH3)		EVQLVESGGGLVQPGGSLRLSCAASGIDFSTYGISWVVRQAPGKGLEWIAIYIPDYGITDY. WVNGRFTISLDNAQNTVYLMNSLRAEDTAVYYCARSGSYYSRSGWGAHYFNLWGQGT TVSS
73	VH16-19-B11-sc01 (VH3)		EVQLVESGGGLVQPGGSLRLSCAASGIDFSTYGISWVVRQAPGKGLEWIGIYIPDYGITDY. WVNGRFTISRDNKNTVYLMNSLRAEDTAVYYCARSGSYYSRSGWGAHYFNLWGQGT TVSS
74	VH16-19-B11-sc02 (VH3)		EVQLVESGGGLVQPGGSLRLSCKASGIDFSTYGISWVVRQAPGKGLEWIAIYIPDYGITDY. WVNGRFTISLDNSKNTVYLMNSLRAEDTAVYYCARSGSYYSRSGWGAHYFNLWGQGT TVSS
75	VH16-19-B11-sc10 or sc11 (VH3)		EVQLVESGGGLVQPGGSLRLSCAASGIDFSTYGISWVVRQAPGKGLEWIAIYIPDYGITDY. WVNGRFTISLDNAQNTVYLMNSLRAEDTAVYYCARSGSYYSRSGWGAHYFNLWGQGT TVSS
76	LCDR1 (L24-L42; AHo numbering)		QASESISSWLA
77	LCDR2 (L58-L72; AHo numbering)		KASTLAS
78	LCDR3 (L107-L138; AHo numbering)		QGYILDSSVDDNV
79	LCDR1 (Kabat)		QASESISSWLA
80	LCDR2 (Kabat)		KASTLAS
81	LCDR3		QGYILDSSVDDNV

	(Kabat)		
82	LCDR1 (Chothia)	SESISSW	
83	LCDR2 (Chothia)	KAS	
84	LCDR3 (Chothia)	YYLDSSVDDN	
85	VL16-19-B11-sc06 (Vk1-sk12)	DIQMTQSPSSLSASVGDRVTINCAQASESISSWLAWYQQKPGKRPKLLIYKASTLASGVPSR GSGGTDYTLTISSLPEDFATYTCQGYLDSSVDDNVFEGGTKLTVLG	
86	VL16-19-B11-sc01 (Vk1-sk12)	DIQMTQSPSSLSASVGDRVTITCAQASESISSWLAWYQQKPGKAPKLLIYKASTLASGVPSR GSGGTDFTLTISSLQPEDFATYTCQGYLDSSVDDNVFEGGTKLTVLG	
87	VL16-19-B11-sc02 (Vk1-sk12)	DIQMTQSPSSLSASVGDRVTINCAQASESISSWLAWYQQKPGKAPKLLIYKASTLASGVPSR GSGGTEFTLTISGLQPADFATYTCQGYLDSSVDDNVFEGGTKLTVLG	
88	VL16-19-B11-sc10 (Vk1-sk12)	DIQMTQSPSSLSASVGDRVTINCAQASESISSWLAWYQQKPGKRPKLLIYKASTLASGVPSR GSGGTDYTLTISSLPEDFATYTCQGYLDSSVDDNVFEGTGTGTVLG	
89	VL16-19-B11-sc11 (Vk1-sk12)	DIQMTQSPSSLSASVGDRVTINCAQASESISSWLAWYQQKPGQRPKLLIYKASTLASGVPSR GSGGTDYTLTISSLPEDFATYTCQGYLDSSVDDNVFEGTGTGTVLG	
Anti-HSA domain (19-01-H04-sc03)			
90	HCDR1 (H27-H42; AHo numbering)	GFSLSNAMG	
91	HCDR2 (H57-H76; AHo numbering)	IISVGGFTYYASWAKG	
92	HCDR3 (H108-H138; AHo numbering)	RDRHGGDSSGAFYL	
93	HCDR1 (Kabat)	SNAMG	
94	HCDR2 (Kabat)	IISVGGFTYYASWAKG	
95	HCDR3	DRHGGDSSGAFYL	

	(Kabat)		
96	HCDR1 (Chothia)		GFSLSSN
97	HCDR2 (Chothia)		VGG
98	HCDR3 (Chothia)		RHGGDSSGAFY
99	VH		EVQLVESGGGLVQPGGSLRLSCAASGFSLSNMGWVVRQAPGKGLYIGIISVGGFTYYA WAKGRFTISRDNKNTVYLMNSLRAEDTATYFCARDRHHGGDSSGAFYLVGGGTLVTV
100	LCDR1 (L24-L42; AHo numbering)		QSSESVYSNNQLS
101	LCDR2 (L58-L72; AHo numbering)		DASDLAS
102	LCDR3 (L107-L138; AHo numbering)		AGGFSSSDTA
103	LCDR1 (Kabat)		QSSESVYSNNQLS
104	LCDR2 (Kabat)		DASDLAS
105	LCDR3 (Kabat)		AGGFSSSDTA
106	LCDR1 (Chothia)		SESVYSNNQ
107	LCDR2 (Chothia)		DAS
108	LCDR3 (Chothia)		GFSSSDT
109	VL		DIQMTQSPSSLSASVGDRVTITCQSSESVYSNNQLSWYQQKPGQPPKLLIYDASDLASGVI FSGSGGTDFLTLSLQPEDFATYCAAGFSSSDTAFGGGTKLTVLG

110	scFv	DIQMTQSPSSLSASVGDRVTITCQSSSESVYNNQLSWYQQKPGQPPKLLIYDASDLASGVI FSGSGGTDFLTITSLQPEDFATYCAAGFSSSDTAFGGGTKLTVLGGGGSGGGSGG GSGGGSEVQLVESGGGLVQPGGSLRLSCAASGFSLSNAMGWVRQAPGKGLIYIGISV GFTYYASWAKGRFTISRDNSKNTVYLQMNSLR AEDTATYFCARDRHGGDSSGAFYLVWG TLVTVSS
Anti-HSA domain (23-13-A01-sc03)		
111	HCDR1 (H27-H42; AHo numbering)	GFSFSSSYWIC
112	HCDR2 (H57-H76; AHo numbering)	CVFTGDDGTTYASWAKG
113	HCDR3 (H108-H138; AHo numbering)	RPVSVVYYGMDL
114	HCDR1 (Kabat)	SSYWIC
115	HCDR2 (Kabat)	CVFTGDDGTTYASWAKG
116	HCDR3 (Kabat)	PVSVVYYGMDL
117	HCDR1 (Chothia)	GFSFSSSY
118	HCDR2 (Chothia)	TGDG
119	HCDR3 (Chothia)	VSVVYYGMD
120	VH	EVQLVESGGGLVQPGGSLRLSCAASGFSFSSSYWICWVRQAPGKGLWVGCVFTGDDGTI YASWAKGRFTISRDNSKNTVYLQMNSLR AEDTATYFCARPVSVVYYGMDLWGGGTLV SS
121	LCDR1 (L24-L42; AHo numbering)	QASQIISRSA

122	LCDR2 (L58-L72; AHo numbering)	QASKLAS
123	LCDR3 (L107-L138; AHo numbering)	QCTYIDSNFGA
124	LCDR1 (Kabat)	QASQIISRSA
125	LCDR2 (Kabat)	QASKLAS
126	LCDR3 (Kabat)	QCTYIDSNFGA
127	LCDR1 (Chothia)	SQIISR
128	LCDR2 (Chothia)	QAS
129	LCDR3 (Chothia)	TYIDSNFG
130	VL	DVVMQTQSPSSLASVGDRTTTCQASQIISRSAWYQQKPGQPPKLLIYQASKLASGVPSR GSGGTDFTLTISLQPEDFATYYCQCTYIDSNFGAFGGGKLTVLG
131	scFv	DVVMQTQSPSSLASVGDRTTTCQASQIISRSAWYQQKPGQPPKLLIYQASKLASGVPSR GSGGTDFTLTISLQPEDFATYYCQCTYIDSNFGAFGGGKLTVLGGGGSGGGSGGG GGGSEVQLVESGGGLVQPGGSLRLSCAASGFSFSSSYWICWVRQAPGKGLEWVGCVF1 DGTYYASWAKGRFTISRDNKNTVYLQMNSLR AEDTATYFCARPVSVYYGMDLWG1 TLVTVSS
Additional Linkers		
132	Linker sequence unit	GGGGS
133	Generic linker sequence	(GmS) ⁿ , with m being selected from 2, 3 and 4 and with n being selected from 2, 3, 4, 5 and 6
TNFα		
134	Transmembrane TNF α	MSTESMIRDVELAEALPKKTTGGPQGSRRCLFLSLFSFLIVAGATTLCLLHFGVIGPQREI RDLSLISPLAQAVRSSSRTPSDKPV AHV V ANPQAEGQLQWLNRRANALLANGVELRDNC

		VPSEGLYLIYSQVLFKGGQCPSTHVLLTHTISRIVSYQTKVNLLSAIKSPCQRETPEGAEA WYEPYILGGVFLQLEKGDRLSAEINRPDYLDFAESGQVYFGIAL
135	Soluble TNF α	VRSSSRTPSDKPVAVHVVANPQAEGLQWLNRANALLANGVELRDNQLVVPSEGLYLIY QVLFKGGQCPSTHVLLTHTISRIVSYQTKVNLLSAIKSPCQRETPEGAEAKPWYEPYILG FQLEKGDRLSAEINRPDYLDFAESGQVYFGIAL
Examples of multispecific molecules of the present invention		
136	A3 LC	DIQMTQSPSSLSASVGDRVITITCQSSSEVYSNNQLSWYQQKPGQPPLLIIYDASDLASGVI FSGSGGTDFLTITSLQPEDFATYYCAGGFSSSDTAAGGGTKLTVLGRTRTVAAPSVFII SDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSYLSSTL SKADYEKHKVYACEVTHQGLSSPVTKSFNRGECGGGGGGGGDIQMTQSPSSLSASVG VTINCQASESISWLAWYQQKPKRPPKLLIYKASTLASGVPSRFSGSGGTDYTLTISLQI DFATYYCQGYILDSSVDNVEFGGTKLTVLGGGGGGGGGGGGGGGGGGSEVQLVES GGLVQPGGSLRLSCAASGIDFSTYGISWVRQAPGKGLEWIAIYIPDYGITDYASWVNGRF SLDNAQNTVYVYLMNSLRAEDTAVYYCARSGSYYSRGGWGAHYFNLWGQGTLLVTVSS
137	A3 HC	EVQLVESGGGLVQPGGSLRLSCAASGFSLSNAMGWVRQAPGKGLYIGIISVGGFTYYA WAKGRFTISRDNKNTVYVYLMNSLRAEDTATYFCARDRHGGDSSGAFYLVWGQGTLLVTV ASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSS YLSNVVTPSSSLGTQYICNVNHHKPSNTKVDKKVEPKSCGGGGGGGGGGGGGGGGGG SASVGDRVITITCQASQTINYWLAWYQQKPKAPKLLIYRSTLASGVPSRFSGSGGTD TISLQPEDFATYYCQNYYYSTSRSYFGGGTKLTVLGGGGGGGGGGGGGGGGGGGGSE LVESGGGLVQPGGSLRLSCAASGFSFSDYWMCWVRQAPGKLEWIGCIYAGDVDDTD SWARGRTISRDNKNTVYVYLMNSLRAEDTAVYYCARVDGFDITYFDLWGQGTLLVTVS DIQMTQSPSSLSASVGDRVITITCQASQTINYWLAWYQQKPKAPKLLIYRSTLASGVPS SGSGGTDFLTITSLQPEDFATYYCQNYYYSTSRSYFGGGTKLTVLGRTRTVAAPSVFI PSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSYLSST LSKADYEKHKVYACEVTHQGLSSPVTKSFNRGECGGGGGGGGGGGGGGGGGGGGGG RVTINCQASESISWLAWYQQKPKRPPKLLIYKASTLASGVPSRFSGSGGTDYTLTISL EDFATYYCQGYILDSSVDNVEFGGTKLTVLGGGGGGGGGGGGGGGGGGGGGGGGGGV GGGLVQPGGSLRLSCAASGIDFSTYGISWVRQAPGKLEWIAIYIPDYGITDYASWVNGF ISLDNAQNTVYVYLMNSLRAEDTAVYYCARSGSYYSRGGWGAHYFNLWGQGTLLVTVSS
138	A4 LC	DIQMTQSPSSLSASVGDRVITITCQASQTINYWLAWYQQKPKAPKLLIYRSTLASGVPS SGSGGTDFLTITSLQPEDFATYYCQNYYYSTSRSYFGGGTKLTVLGRTRTVAAPSVFI PSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSYLSST LSKADYEKHKVYACEVTHQGLSSPVTKSFNRGECGGGGGGGGGGGGGGGGGGGGGG RVTINCQASESISWLAWYQQKPKRPPKLLIYKASTLASGVPSRFSGSGGTDYTLTISL EDFATYYCQGYILDSSVDNVEFGGTKLTVLGGGGGGGGGGGGGGGGGGGGGGGGGGV GGGLVQPGGSLRLSCAASGIDFSTYGISWVRQAPGKLEWIAIYIPDYGITDYASWVNGF ISLDNAQNTVYVYLMNSLRAEDTAVYYCARSGSYYSRGGWGAHYFNLWGQGTLLVTVSS
139	A4 HC	EVQLVESGGGLVQPGGSLRLSCAASGFSFSDYWMCWVRQAPGKGLWIGCIYAGDVDD WAKGRFTISRDNKNTVYVYLMNSLRAEDTATYFCARDRHGGDSSGAFYLVWGQGTLLVTV

		DYASWARGRFTISRDN SKNTVYLQMNSLRAEDTAVYYCARVDGFDITYFDLWGQGTLV SSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQ GLYSLSSVTVPSSSLGTQTYICNVNHKPSNTKVDKKVEPKSCGGGGGGGGSDIQMTQS SLSASVGDRVTITCQSSSEVYSNNQLSWYQQKPGQPPKLLIYDASDLASGVPSRFSGSGC DFTLTISSLQPEDFATYYCAGGFSSSDTAFGGGKLTVLGGGGGGGGGGGGGGGGG VQLVESGGGLVQPGGSLRLSCAASGFSLSNNAMGWVRQAPGKGLYIGHSVGGFTYYAS AKGRFTISRDN SKNTVYLQMNSLRAEDTATYFCARDRHGGDSSGAFYLWGQGTLVTVSS DIQMTQSPSSLSASVGDRVTINCAQASESISWLA WYQQKPKRPPKLLIYKASTLASGVPSR GSGGTDYTLTISSLQPEDFATYYCQGYLLDSSVDDNVFEGGKLTVLGRTRTVAAPSVF PPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSITYSLSS TLKADYEEKHKVYACEVTHQGLSSPVTKSFNRGECGGGGGGGGSDIQMTQSPSSLSAS DRVITTCQSSSEVYSNNQLSWYQQKPGQPPKLLIYDASDLASGVPSRFSGSGTDFTLTI LQPEDFATYYCAGGFSSSDTAFGGGKLTVLGGGGGGGGGGGGGGGGGGGGGGGGGG GGGLVQPGGSLRLSCAASGFSLSNNAMGWVRQAPGKGLYIGHSVGGFTYYASWAKGR SRDN SKNTVYLQMNSLRAEDTATYFCARDRHGGDSSGAFYLWGQGTLVTVSS
140	A5 LC	EVQLVESGGGLVQPGGSLRLSCAASGIDFSTYGISWVRQAPGKGLEWIAIYIPDYGITDY WVNGRFTISLDNAQNTVYLQMNSLRAEDTAVYYCARSGSYYSRGGWGAHYFNLWGQGT TVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVI SSGLYSLSSVTVPSSSLGTQTYICNVNHKPSNTKVDKKVEPKSCGGGGGGGGSDIQMT PSSLSASVGDRVTITCQASQTINYLAWYQQKPKAPKLLIYRASTLASGVPSRFSGSGC DFTLTISSLQPEDFATYYCQNYYSYTSRSYGFGGKLTVLGGGGGGGGGGGGGGGGG SEVQLVESGGGLVQPGGSLRLSCAASGFSFSSDYWMCWVRQAPGKGLEWIGCIYAGDVI TDYASWARGRFTISRDN SKNTVYLQMNSLRAEDTAVYYCARVDGFDITYFDLWGQGTLV VSS
141	A5 HC	DIQMTQSPSSLSASVGDRVTITCQASQTINYLAWYQQKPKAPKLLIYRASTLASGVPS SGSGGTDFTLTISSLQPEDFATYYCQNYYSYTSRSYGFGGKLTVLGGGGGGGGGGGGGG GGGLVQPGGSLRLSCAASGFSLSNNAMGWVRQAPGKGLYIGHSVGGFTYYASWAKGRF RDNSKNTVYLQMNSLRAEDTATYFCARDRHGGDSSGAFYLWGQGTLVTVSSGGGGGGG GGGGGGGGGGSDIQMTQSPSSLSASVGDRVTITCQSSSEVYSNNQLSWYQQKPGQPPKI YDASDLASGVPSRFSGSGTDFTLTISSLQPEDFATYYCAGGFSSSDTAFGGGKLTVL GGGSEVQLVESGGGLVQPGGSLRLSCAASGFSFSSDYWMCWVRQAPGKGLEWIGCIYAC
142	A6	

		VDDTDYASWARGRFTISRDNKNTVYLQMNSLRAEDTAVYYCARVDGFDITYFDLWGC LVTVSSGGGGGGSDIQMTQSPSSLASVGDRTVINCQASESISSWLAWYQQKPKGRF LIYKASTLASGVPSRFSGSGTDYTLTISSLQPEDFATYQCQGYLDSSVDDNVFGGGTk VLGGGGGGGGGGGGGGGGGGVQPGGSLRLSCAASGIDFSTYGISW QAPGKGLEWIAIYIPDYGITDYASWVNGRFTISLDNAQNTVYLQMNSLRAEDTAVYYC/ SGSYYSRGWGAHYFNLWGQGTLVTVSS
143	A7	DIQMTQSPSSLASVGDRTVINCQASESISSWLAWYQQKPKRPKLLIYKASTLASGVPSR GSGGTDYTLTISSLQPEDFATYQCQGYLDSSVDDNVFGGGTkTLVLGGGGGGSEVQLV GGGLVQPGGSLRLSCAASGFSSESDYWMCWVRQAPGKGLEWIGCIYAGVDDTDYASW RGRFTISRDNKNTVYLQMNSLRAEDTAVYYCARVDGFDITYFDLWGQGTLVTVSSGGC SGGGGGGGGGGGSDIQMTQSPSSLASVGDRTVITCQASQTINYWLAWYQQKPKGA LLIYRASTLASGVPSRFSGSGTDFTLTISSLQPEDFATYQCQNYYSTSRSYGFGGGTKI VLGGGGGGSEVQLVESGGGLVQPGGSLRLSCAASGIDFSTYGISWVRQAPGKGLEWIAIY YGITDYASWVNGRFTISLDNAQNTVYLQMNSLRAEDTAVYYCARSGSYYSRGWGAHYF WGQGTLVTVSSGGGGGGGGDIQMTQSPSSLASVGDRTVITCQSSSVYNNQLSWYI KPGQPKLLIYDASDLASGVPSRFSGSGTDFTLTISSLQPEDFATYACAGFSSSDTAF GTKLTVLGGGGGGGGGGGGGGGGGGVQPGGSLRLSCAASGFSLS MGWVRQAPGKGLEIYIGISVGGFTYASWAKGRFTISRDNKNTVYLQMNSLRAEDTAT CARDRHGGDSSGAFYLVWGQGTLVTVSS
144	A8	DIQMTQSPSSLASVGDRTVITCQSSSVYNNQLSWYQQKPGQPPKLLIYDASDLASGVI FSGSGGTDFTLTISSLQPEDFATYACAGFSSSDTAFGGGTkTLVLGGGGGGSEVQLVES GLVQPGGSLRLSCAASGIDFSTYGISWVRQAPGKGLEWIAIYIPDYGITDYASWVNGRFT LDNAQNTVYLQMNSLRAEDTAVYYCARSGSYYSRGWGAHYFNLWGQGTLVTVSSGGC GGGGGGGGGGGGSDIQMTQSPSSLASVGDRTVINCQASESISSWLAWYQQKPKRPI LIYKASTLASGVPSRFSGSGTDYTLTISSLQPEDFATYQCQGYLDSSVDDNVFGGGTk VLGGGGGGSEVQLVESGGGLVQPGGSLRLSCAASGFSLSNMGWVRQAPGKGLEIYIGIS GGFTYASWAKGRFTISRDNKNTVYLQMNSLRAEDTATYFCARDRHGGDSSGAFYLV GTLVTVSSGGGGGGGGDIQMTQSPSSLASVGDRTVITCQASQTINYWLAWYQQKPG PKLLIYRASTLASGVPSRFSGSGTDFTLTISSLQPEDFATYQCQNYYSTSRSYGFGGGT LTVLGGGGGGGGGGGGGGGGGGVQPGGSLRLSCAASGFSFSDYW CWVRQAPGKGLEWIGCIYAGVDDTDYASWARGRFTISRDNKNTVYLQMNSLRAEDT

145	A9	VYYCARVDGFDITYFDLWGQGTLVTVSS DIQMTQSPSSLSASVGDRVTINCAASEISSWLAWYQQKPKRPKLLIYKASTLASGVPSR GSGGTDYTLTISGLQPEDFATYYCQGYLDSSVDNDFGTGKTVTLGGGGSEVQLV GGGLVQPGGSLRLSCAASGFSFSDYWMCWVVRQAPGKGLEWIGCIYAGVDVDDTDYASW RGRFTISRDNKNTVYLMNSLRAEDTAVYYCARVDGFDITYFDLWGQGTLVTVSSGGC SGGGSGGGGGGGDIQMTQSPSSLSASVGDRVTITCQASQTINYLAWYQQKPKGA LLIYRASTLASGVPSRFSGSGTDFTLTISLQPEDFATYYCQNYYYSTSRSYGFGGGTKI VLGGGGSEVQLVESGGGLVQPGGSLRLSCAASGIDFSTYGISWVRQAPGKGLEWIAIYI YGITDYASWVNGRFTISLDNAQNTVYLMNSLRAEDTAVYYCARSGSYYSRGGWAHYF WGQGTLVTVSSGGGGGGGGDIQMTQSPSSLSASVGDRVTITCQSSSVYNNQLSWYI KPGQPPKLLIYDASDLASGVPSRFSGSGTDFTLTISLQPEDFATYYCAGGFSSTDAF GTKLTVLGGGGGGGGGGGGGGGGGGVQLVESGGGLVQPGGSLRLSCAASGFSLS MGWVRQAPGKGLEIYIGISVGGFTYYASWAKGRFTISRDNKNTVYLMNSLRAEDTAT CARDRHGGDSSGAFYLVGQGTLVTVSS
146	A10	DIQMTQSPSSLSASVGDRVTINCAASEISSWLAWYQQKPKRPKLLIYKASTLASGVPSR GSGGTDYTLTISLQPEDFATYYCQGYLDSSVDNDFGTGKTVTLGGGGSEVQLV GGGLVQPGGSLRLSCAASGFSFSDYWMCWVVRQAPGKGLEWIGCIYAGVDVDDTDYASW RGRFTISRDNKNTVYLMNSLRAEDTAVYYCARVDGFDITYFDLWGQGTLVTVSSGGC SGGGSGGGGGGGDIQMTQSPSSLSASVGDRVTITCQASQTINYLAWYQQKPKGA LLIYRASTLASGVPSRFSGSGTDFTLTISLQPEDFATYYCQNYYYSTSRSYGFGGGTKI VLGGGGSEVQLVESGGGLVQPGGSLRLSCAASGIDFSTYGISWVRQAPGKGLEWIAIYI YGITDYASWVNGRFTISLDNAQNTVYLMNSLRAEDTAVYYCARSGSYYSRGGWAHYF WGQGTLVTVSSGGGGGGGGDIQMTQSPSSLSASVGDRVTITCQSSSVYNNQLSWYI KPGQPPKLLIYDASDLASGVPSRFSGSGTDFTLTISLQPEDFATYYCAGGFSSTDAF GTKLTVLGGGGGGGGGGGGGGGGGGVQLVESGGGLVQPGGSLRLSCAASGFSLS MGWVRQAPGKGLEIYIGISVGGFTYYASWAKGRFTISRDNKNTVYLMNSLRAEDTAT CARDRHGGDSSGAFYLVGQGTLVTVSS
147	A11	DIQMTQSPSSLSASVGDRVTINCAASEISSWLAWYQQKPKRPKLLIYKASTLASGVPSR GSGGTDYTLTISGLQPEDFATYYCQGYLDSSVDNDFGTGKTVTLGGGGSEVQLV GGGLVQPGGSLRLSCAASGFSFSDYWMCWVVRQAPGKGLEWIGCIYAGVDVDDTDYASW RGRFTISRDNKNTVYLMNSLRAEDTAVYYCARVDGFDITYFDLWGQGTLVTVSSGGC

		SGGGSGGGSGGGSIQMTQSPSSLSASVGDRVTITCQASQTINYWLAWYQQKPGKAP LIYRASTLASGVPSRFSGSGTDFLTIISSLQPEDFATYYCQNYYSTSRSYGFGGTKL. LGGGGSEVQLVESGGGLVQPGGSLRLSCAASGIDFSTYGISWVRQAPGKGLEWIAIYP GITDYASWVNGRFTISLDNAQNTVYLQMNSLRAREDATVYCARSGSYSRGWGAHYFN WGQGTLVTVSSGGGGGGGSIQMTQSPSSLSASVGDRVTTTCQSSES VYSNNQLSWYQ PGPPKLLIYDASDLASGVPSRFSGSGTDFLTIISSLQPEDFATYYCAGGFSSSDTAFG TKLTVLGGGGGGGGGGGGGGSEVQLVESGGGLVQPGGSLRLSCAASGFSLSN MGWVRQAPGKLEYIGHSVGGTTYASWAKGRFTISRDNKNTVYLQMNSLRAREDAT CARDRHGGDSGAFYLVGGQTLVTVSS
148	A12	DIQMTQSPSSLSASVGDRVTTNCQASEISSWLAWYQQKPGQRPKLLIKASTLASGVPSR GSGGTDTYLTIISSLQPEDFATYYCQGYLYDSSVDNVFGTGTKVTLVGGGGSEVQLV GGGLVQPGGSLRLSCAASGFSSDYWMCWVRQAPGKGLEWIGCIYAGDVDDTDYASW RGRTISRDNKNTVYLQMNSLRAREDATVYCARVDGFDITYFDLWGQGTIVTVSSGGC SGGGSGGGGGGGSIQMTQSPSSLSASVGDRVTTITCQASQTINYWLAWYQQKPGKAP LIYRASTLASGVPSRFSGSGTDFLTIISSLQPEDFATYYCQNYYSTSRSYGFGGTKL. LGGGGSEVQLVESGGGLVQPGGSLRLSCAASGIDFSTYGISWVRQAPGKGLEWIAIYP GITDYASWVNGRFTISLDNAQNTVYLQMNSLRAREDATVYCARSGSYSRGWGAHYFN WGQGTLVTVSSGGGGGGGSIQMTQSPSSLSASVGDRVTTITCQSSES VYSNNQLSWYQ PGPPKLLIYDASDLASGVPSRFSGSGTDFLTIISSLQPEDFATYYCAGGFSSSDTAFG TKLTVLGGGGGGGGGGGGGGSEVQLVESGGGLVQPGGSLRLSCAASGFSLSN MGWVRQAPGKLEYIGHSVGGTTYASWAKGRFTISRDNKNTVYLQMNSLRAREDAT CARDRHGGDSGAFYLVGGQTLVTVSS
149	A13	DIQMTQSPSSLSASVGDRVTTNCQASEISSWLAWYQQKPGKRPKLLIKASTLASGVPSR GSGGTDTYLTIISSLQPEDFATYYCQGYLYDSSVDNVFGGTGKTIVLGGGGSEVQLV GGGLVQPGGSLRLSCAASGFSLSNAMGWVRQAPGKLEYIGHSVGGTTYASWAKGR SRDNKNTVYLQMNSLRAREDATATFCARDRHGGDSGAFYLVGGQTLVTVSSGGGGSC GGGGGGGGGGDIQMTQSPSSLSASVGDRVTTITCQSSES VYSNNQLSWYQQKPGQPPK IYDASDLASGVPSRFSGSGTDFLTIISSLQPEDFATYYCAGGFSSSDTAFFGGGTCLTVI GGGGSEVQLVESGGGLVQPGGSLRLSCAASGIDFSTYGISWVRQAPGKGLEWIAIYTPDY TDYASWVNGRFTISLDNAQNTVYLQMNSLRAREDATVYCARSGSYSRGWGAHYFN QGTLVTVSS

It is appreciated that certain features of the invention, which are, for clarity, described in the context of separate embodiments, may also be provided in combination in a single embodiment. Conversely, various features of the invention, which are, for brevity, described in the context of a single embodiment, may also be provided separately or in any suitable sub-combination. All combinations of the embodiments pertaining to the invention are specifically embraced by the present invention and are disclosed herein just as if each and every combination was individually and explicitly disclosed. In addition, all sub-combinations of the various embodiments and elements thereof are also specifically embraced by the present invention and are disclosed herein just as if each and every such sub-combination was individually and explicitly disclosed herein.

The present invention is not to be limited in scope by the specific embodiments described herein. Indeed, various modifications of the invention in addition to those described herein will become apparent to those skilled in the art from the foregoing description. Such modifications are intended to fall within the scope of the appended claims.

To the extent possible under the respective patent law, all patents, applications, publications, test methods, literature, and other materials cited herein are hereby incorporated by reference.

The following Examples illustrates the invention described above, but is not, however, intended to limit the scope of the invention in any way. Other test models known as such to the person skilled in the pertinent art can also determine the beneficial effects of the claimed invention.

EXAMPLES

Example 1: Generation of rabbit antibodies directed against human IL-17A

1.1 Immunization

Rabbits have been immunized with recombinantly produced and purified IL-17A (Peprotech, Cat. No. 200-17). During the course of the immunization, the strength of the

humoral immune response against the antigen was qualitatively assessed by determining the maximal dilution (titer) for the serum of each rabbit that still results in detectable binding of the polyclonal serum antibodies to the antigen. Serum antibody titers against the immobilized antigen (recombinant human IL-17A) were assessed using an enzyme-linked immunosorbent
5 assay (ELISA).

The ability of the rabbit sera to inhibit the biological activity of IL-17A was assessed using a cell-based assay with HT-29 cells that secrete GRO- α upon IL-17A-stimulation. 50'000 HT-29 cells per well were seeded in 96-well plates and stimulated with 50 ng/ml IL-17A. 5-fold serial dilutions of final bleeds of IL-17A immunized rabbits were tested for the
10 potency to neutralize biological function of IL-17A. All five sera inhibited GRO- α secretion of HT-29 cells. Lymphocytes were isolated from spleens of immunized animals and were a subject for the subsequent Hit Identification procedures.

1.2 Identification of Anti-IL-17A Antibodies

1.2.1 Isolation of B Cells Expressing Anti-IL-17A Antibodies

15 To identify IL-17A binding B-cells, IL-17A was labeled with R-Phycoerythrin (RPE). Since the IL-17 receptor A binding site on the labeled IL-17A could potentially be blocked by the bulky RPE label, accessibility of the epitope was confirmed by two approaches. In a first assay, binding of RPE labeled IL-17A to secukinumab and IL-17 receptor A was analyzed using flow-cytometry. Secukinumab and IL-17 receptor A extracellular domain fused to the
20 Fc part of a human IgG1 were captured on protein G beads, and binding of RPE labeled IL-17A was confirmed by flow-cytometry. Binding of IL-17A to secukinumab and IL-17 receptor A Fc chimera was thereby confirmed while no binding to an unrelated IgG or cytokine receptor was detected. In a second approach, biological activity of the labeled IL-17A was confirmed in the HT-29 assay. As shown in FIG. 1, RPE labeled IL-17A showed
25 only slightly reduced biological activity when compared to unlabeled IL-17A (1.5-fold higher EC₅₀ for induction of IL-17A-dependent GRO- α secretion). Therefore, it was confirmed that the labeled IL-17A is suitable for use in the sorting process.

1.2.2 Screening of IL-17A-Binding Supernatants

The results obtained during the screening phase are based on assays performed with non-purified antibodies from culture supernatants of antibody secreting cells (ASC). ASC supernatants were screened in a high-throughput ELISA for binding to recombinant human IL-17A. IL-17A-binding supernatants were further characterized for their binding kinetics and for their potential to neutralize the biological activity of IL-17A in the cell-based HT-29 assay and a competition ELISA. IL-17A-binding supernatants were further characterized for binding to Cynomolgus monkey IL-17A by ELISA.

1.2.2.1 IL-17A binding by ELISA

To identify B-cell clones that produce antibodies binding human IL-17A, cell culture supernatants of B-cell clones were analyzed for the presence of antibodies to human IL-17A by ELISA. The ELISA method used assesses the “quantity” of antibodies of the IgG subtype bound to recombinant human IL-17A, gives however no information about the affinity or the concentration of the antibodies.

1.2.2.2 Affinity to human IL-17A

Binding affinities of monoclonal rabbit antibodies toward human IL-17A from culture supernatants that qualified positive during the primary screening were determined by surface plasmon resonance (SPR) using a MASS-1 SPR device (Sierra Sensors). For affinity screening, an antibody specific for the Fc region of rabbit IgGs was immobilized on a sensor chip (SPR-2 Affinity Sensor, High Capacity Amine, Sierra Sensors) using a standard amine-coupling procedure. Rabbit monoclonal antibodies in B-cell supernatants were captured by the immobilized anti-rabbit IgG antibody. A minimal IgG concentration in the B-cell supernatants is required to allow sufficient capture. After capturing of the monoclonal antibodies, human IL-17A was injected into the flow cells for 3 min at a concentration of 90 nM, and dissociation of the protein from the IgG captured on the sensor chip was allowed to proceed for 5 min. The apparent dissociation (k_d) and association (k_a) rate constants and the

apparent dissociation equilibrium constant (K_D) were calculated with the MASS-1 analysis software (Analyzer, Sierra Sensors) using one-to-one Langmuir binding.

1.2.2.3 Neutralization of IL-17A by the HT-29 assay and a competition ELISA

For the assessment of potency, a cell-based assay (HT-29 assay), as well as a receptor
5 ligand competition-ELISA, were developed.

The ability of the antibodies in B cell supernatant to inhibit the biological activity of IL-17A was assessed using a cell-based assay with HT-29 cells that secrete GRO- α upon IL-17A stimulation. 50'000 HT-29 cells per well were seeded in 96-well plates and stimulated with 5 ng/ml IL-17A. B cell supernatants at a final concentration of 50% were analyzed for the
10 potency to neutralize biological function of IL-17A.

The inhibition of hIL-17A binding to hIL-17RA was assessed by competitive ELISA. hIL-17RA was coated on the ELISA plate at a concentration of 4 μ g/ml. Biotinylated hIL-17A (20 ng/ml) was preincubated with B cell supernatants (95%) for 1 h, the mixture was then added to the ELISA plate to allow binding to the hIL-17RA for 1.5 h. Then, Streptavidin-
15 HRP, used to Biotinylated IL-17A was detected by Streptavidin-HRP.

1.2.2.4 Species cross-reactivity (binding to Cynomolgus monkey IL-17A by SPR)

Selected hits were analyzed for species cross-reactivity to cynomolgus monkey IL-17A by SPR. Binding affinities were determined by surface plasmon resonance (SPR) using a MASS-1 SPR device (Sierra Sensors) similarly as described in section 1.2.2.2 for human IL-
20 17A with the exception that 90 nM cynomolgus monkey IL-17A was used instead of human IL-17A.

1.2.2.5 IL-17F binding by ELISA

To confirm specificity of the anti-IL-17A scFvs, binding to all the IL-17 family members (IL-17B, IL-17C, IL-17D, IL-17F and IL-17E) was assessed for the best performing
25 humanized scFvs. The sort supernatants were screened in ELISA for binding to IL-17F

because IL-17F shows the highest identity to IL-17A (47%, see TABLE 2). Binding was quantified relative to the signal obtained with 2 µg/ml secukinumab.

TABLE 2. Identity between IL-17A and the other members of the IL-17 family. Data was obtained by aligning the IL-17A sequence (UniProtKB/Swiss-Prot: Q16552.1) to IL-17B, IL-17C, IL-17D, IL-17F and IL-17E using EMBOSS NEEDLE (with std settings).

IL-17 Family Member	Identity to IL-17A	Similarity to IL-17A
IL-17B	26.6	36.4
IL-17C	23.4	31.8
IL-17D	21.8	27.5
IL-17E	21.3	31.7
IL-17F	47.2	62

1.3 Functional Characterization

Based on pharmacologic properties of monoclonal antibodies in B-cell supernatant a number of clones were selected for hit confirmation analysis. Pharmacologic properties of monoclonal antibodies of the selected clone in B-cell supernatant are presented in TABLE 3.

1.3.1 IL-17A Binding Kinetics (by SPR)

Binding kinetics of a number of purified monoclonal rabbit antibodies to human IL-17A were determined by SPR (Mass-1) measurements. Each IgG was captured via an anti-rabbit IgG (Bethyl Laboratories, Cat. No. A120-111A) coupled to a carboxymethylated dextran surface (SPR-2 Affinity Sensor, High Capacity Amine, Sierra Sensors) and an IL-17A dose response was measured to allow accurate fitting of kinetic parameters. After capturing of the monoclonal antibodies, human IL-17A (Peprotech, Cat. No. 200-17) was injected into the flow cells for 3 min and dissociation of the protein from the IgG captured on the sensor chip was allowed to proceed for 5 min. After each injection cycle, surfaces were regenerated with two injections of 10 mM Glycine-HCl. The apparent dissociation (k_d) and association (k_a) rate constants and the apparent dissociation equilibrium constant (K_D) were calculated with the MASS-1 analysis software (Analyzer, Sierra Sensors) using one-to-one Langmuir binding model and quality of the fits was monitored based on relative Chi2 (Chi2 normalized to the

extrapolated maximal binding level of the analyte), which is a measure for the quality of the curve fitting.

High-affinity binding to human IL-17A was confirmed for all antibodies with K_D values ranging down to sub-picomolar affinities (2.87×10^{-10} to below 1.51×10^{-13} M). Monoclonal antibodies of the selected clone 27-07-G02 have a dissociation constant (K_D) of 1.88 pM, k_a for on-rates of $2.44 \times 10^6 \text{ M}^{-1}\text{s}^{-1}$, and k_d for off-rates of $4.58 \times 10^{-6} \text{ s}^{-1}$ as measured by SPR (see TABLE 3). Monoclonal antibodies of the selected clone 27-31-C04 have a dissociation constant (K_D) of less than 0.5 pM, k_a for on-rates of $1.99 \times 10^6 \text{ M}^{-1}\text{s}^{-1}$, and k_d for off-rates less than $1 \times 10^{-6} \text{ s}^{-1}$ as measured by SPR (see TABLE 3).

1.3.2 Cross-reactivity to cynomolgus monkey IL-17A (by SPR)

Species cross-reactivity for cynomolgus monkey IL-17A was determined for the selected purified monoclonal rabbit antibodies by SPR (Mass-1) measurements similarly as described above. Trenzyme company custom-produced cynomolgus monkey IL-17A was used in the assays. Monoclonal antibodies of the selected clone 27-07-G02 had a dissociation constant (K_D) of 0.6 pM, k_a for on-rates of $1.24 \times 10^6 \text{ M}^{-1}\text{s}^{-1}$, and k_d for off-rates of $7.72 \times 10^{-6} \text{ s}^{-1}$ as measured by SPR (see TABLE 3). The ration of $K_{D\text{cynoIL-17A}}/K_{D\text{humanIL-17A}}$ as measured by SPR was determined to be 0.3 (see TABLE 3). Monoclonal antibodies of the selected clone 27-31-C04 had a dissociation constant (K_D) of less than 25 pM, k_a for on-rates of $1.69 \times 10^6 \text{ M}^{-1}\text{s}^{-1}$, and k_d for off-rates $4.22 \times 10^{-5} \text{ s}^{-1}$ as measured by SPR (see TABLE 3). The ration of $K_{D\text{cynoIL-17A}}/K_{D\text{humanIL-17A}}$ as measured by SPR was determined to be more than 5 (see TABLE 3).

1.3.3 Neutralization of human IL-17A (by the HT-29 assay)

The effects of purified rabbit monoclonal antibodies on IL-17A-induced GRO- α secretion of the human colon carcinoma cell line HT-29 was examined. The potency (IC_{50}) to neutralize IL-17A-induced GRO- α secretion (as quantified by a commercial ELISA) was analyzed for serial dilutions of all antibodies and compared to the potency of secukinumab. Data were analyzed using a four-parameter logistic curve fit, and the molar concentration of IL-17A inhibitor required to reduce GRO- α secretion to 50% (IC_{50}) was derived from inhibition

curves. In order to render IC₅₀ values from different assay plates directly comparable to each other, individual IC₅₀ values on each plate were calibrated against the IC₅₀ of the reference molecule secukinumab that was taken along on each plate (relative IC₅₀: IC_{50, secukinumab}/IC_{50, test antibody}). Relative IC₅₀ values were calculated in mass units (ng/ml) of secukinumab and the scFvs.

Neutralization assays can distinguish potencies of target blocking antibodies only if they bind their target with an equilibrium binding constant (K_D) that is higher than the target concentration used in the potency assay ($K_D > \text{target concentration}$). For the HT-29 assay an IL-17A concentration of 20 ng/ml (= 645 pM) was used. Therefore, theoretically, the HT-29 assay can differentiate potencies between IgGs with $K_D > 645 \text{ pM}$. Since all of the IgGs analyzed showed K_D values below 645 pM, potencies between IgGs with different affinities (but similar mechanism of action) cannot be differentiated with this. Thus, the HT-29 assay was further developed to include 50 pg/ml TNF α in order to better resolve potencies. This cytokine acts synergistically with IL-17A to induce GRO- α secretion, and as a result the IL-17A concentration could be reduced to 1 ng/ml (= 32 pM) in the presence of TNF α .

The selected clone 27-07-G02 showed a potency above 50-fold of (see TABLE 3, FIG. 2). More specifically, the clone 27-07-G02 was shown to have a potency to neutralize IL-17A relative to that of secukinumab (relative potency), determined in an HT-29 assay, of 75 for IC₅₀ and 48 for IC₉₀, and wherein said relative potency is the ratio of the IC₅₀ or IC₉₀ value in ng/mL of secukinumab in the HT-29 assay to the IC₅₀ or IC₉₀ value in ng/mL of the monoclonal antibody of the clone 27-07-G02 in the HT-29 assay (see TABLE 3, FIG. 2).

The selected clone 27-31-C04 showed a potency above 100-fold of secukinumab (see TABLE 3, FIG. 2). More specifically, the selected clone 27-31-C04 was shown to have a potency to neutralize IL-17A relative to that of secukinumab (relative potency), determined in an HT-29 assay, of 287 for IC₅₀ and 704 for IC₉₀, and wherein said relative potency is the ratio of the IC₅₀ or IC₉₀ value in ng/mL of secukinumab in the HT-29 assay to the IC₅₀ or IC₉₀ value in ng/mL of the monoclonal antibody of the clone 27-31-C04 in the HT-29 assay (see TABLE 3, FIG. 2).

1.3.4 Blocking of the human IL-17A/IL-17RA interaction (by competitive ELISA)

The inhibition of hIL-17A binding to hIL-17RA was assessed by competitive ELISA. hIL-17RA was coated on the ELISA plate by adding 50 μ l of PBS containing 4 μ g/ml IL-17RA (Sino Biological Cat. No. 10895-H08H) ON at 4°C. Biotinylated hIL-17A was

5 preincubated with the rabbit monoclonal antibodies for 1 h, the mixture was then added to the ELISA plate to allow binding to the hIL-17RA for 1.5 h. Then, 50 μ l of 10 ng/ml streptavidin-polyHRP40 (SDT Cat. No. SP40C), used to detect the biotinylated IL-17A, was added and the plate was incubated for 1 h. Finally, Tetramethylbenzidine solution (KPL, Cat. No. 53-00-00) was added to develop the plate for 5 to 10 minutes and the reaction was stopped with 1 M

10 HCl. The plate was read using a microtiter plate reader (Infinity reader M200 Pro, Tecan) at a wavelength of 450 nm and 570 nm (reference wavelength).

Dose-response curve obtained for the selected rabbit IgG is represented in FIG. 3. IC₅₀ values and IC₉₀ values compared to the reference secukinumab are summarized in TABLE 3. As mentioned in the previous section, neutralization assays can distinguish potencies of

15 target blocking antibodies only if they bind their target with an equilibrium binding constant (K_D) that is higher than the target concentration used in the potency assay ($K_D > \text{target concentration}$). For the HT-29 assay a IL-17A concentration of 32 pM was used while in the IL-17A/IL-17RA inhibition ELISAs a IL-17A concentration of 193 pM was used. Therefore, theoretically, the HT-29 assay can differentiate potencies between IgGs with $K_D > 32$ pM,

20 while the inhibition ELISA can only differentiate potencies between IgGs with $K_D > 193$ pM. Like in the HT-29 assay, clone 27-07-G02 and clone 27-31-C04 showed a higher potency than secukinumab. The clone 27-07-G02 was shown to have a potency to block IL-17A/IL-17RA interaction relative to that of secukinumab (relative potency), determined in an ELISA assay, of 5 for IC₅₀ and 14 for IC₉₀, and wherein said relative potency is the ratio of the

25 IC₅₀ or IC₉₀ value in ng/mL of secukinumab in the ELISA assay to the IC₅₀ or IC₉₀ value in ng/mL of the monoclonal antibody of the clone 27-07-G02 in the ELISA assay. Similarly, the clone 27-31-C04 was shown to have a potency to block IL-17A/IL-17RA interaction relative to that of secukinumab (relative potency), determined in an ELISA assay, of 8 for IC₅₀ and 21 for IC₉₀, (see TABLE 3).

1.3.5 Cross-reactivity to cynomolgus monkey IL-17A (by HT-29 assay)

50'000 HT-29 cells were plated in each well of a 96-well plate. In addition to serial dilutions of rabbit monoclonal antibodies and the internal reference secukinumab, pre-dilutions of human TNF α (50 pg/ml) and human or cynomolgus monkey IL-17A (1 ng/ml), respectively, were added to the HT-29 cells. After 24 h incubation at 37°C and 5% CO₂, supernatants were collected and GRO- α (CXCL1 chemokine) secretion was quantified by ELISA. In order to render IC₅₀ values obtained with cynomolgus IL-17A directly comparable to each other, individual IC₅₀ values on each plate were calibrated against the IC₅₀ obtained with human IL-17A that was taken along on each plate (relative IC₅₀: IC_{50, cynomolgus IL-17A}/IC_{50, human IL-17A}). Potency of the selected clone is presented in TABLE 3. For the clone 27-07-G02 the potency (IC₅₀) to neutralize cynomolgus IL-17A-induced GRO- α secretion was determined to be 0.64 ng/ml, and for the clone 27-31-C04 the potency (IC₅₀) to cynomolgus IL-17A-induced GRO- α secretion was determined to be 0.49 ng/ml (see TABLE 3).

AMENDED DESCRIPTION

TABLE 3. Pharmacodynamic properties of clones 27-07-G02 and 27-31-C04 monoclonal rabbit anti-IL-17A IgGs selected from immunization.

IgG	Affinity for human IL-17A (SPR)			Affinity for cynomolgus IL-17A (SPR)			Ratio IL-17A binding SPR ($KD_{\text{cynolL-17A}}$) /($KD_{\text{human IL-17A}}$)	
	Clone ID	k_a [$M^{-1} s^{-1}$]	k_d [s^{-1}]	K_D [M]	k_a [$M^{-1} s^{-1}$]	k_d [s^{-1}]	K_D [M]	
	27-07-G02	2.44E+06	4.58E-06	1.88E-12	1.24E+06	7.72E-07	6.22E-13	0.3

*: IC_{50} , Secukinumab/ IC_{50} , Test Sample**: IC_{50} , cyno IL17A/ IC_{50} , human IL17A

IgG	Potency in HT-29 assay		Potency in IL17A-IL17RA ELISA		HT-29 assay with human and cyno IL17A			Relative binding to IL-17F (ELISA)
	Clone ID	rel. IC ₅₀ *	rel. IC ₉₀ *	rel. IC ₅₀ *	rel. IC ₉₀ *	IC ₅₀ [ng/mL] human IL17A	IC ₅₀ [ng/mL] cyno IL17A	rel. IC ₅₀ **
27-07-G02	74.88	47.95	4.94	13.64	0.32	0.64	1.98	1%

IgG	Affinity for human IL-17A (SPR)			Affinity for cynomolgus IL-17A (SPR)			Ratio IL-17A binding SPR ($KD_{\text{cynolL-17A}}$) /($KD_{\text{human IL-17A}}$)	
	Clone ID	k_a [$M^{-1} s^{-1}$]	k_d [s^{-1}]	K_D [M]	k_a [$M^{-1} s^{-1}$]	k_d [s^{-1}]		K_D [M]
	27-31-C04	1.99E+06	<1E-06	<5.03E-13	1.69E+06	4.22E-05	2.50E-11	> 5.0

*: IC_{50} , Secukinumab/ IC_{50} , Test Sample**: IC_{50} , cyno IL17A/ IC_{50} , human IL17A

IgG	Potency in HT-29 assay		Potency in IL17A-IL17RA ELISA		HT-29 assay with human and cyno IL17A			Relative binding to IL-17F (ELISA)	
	Clone ID	rel. IC ₅₀ *	rel. IC ₉₀ *	rel. IC ₅₀ *	rel. IC ₉₀ *	IC ₅₀ [ng/mL] human IL17A	IC ₅₀ [ng/mL] cyno IL17A	rel. IC ₅₀ **	Rel. OD _{450-690nm} [OD _{IL17F} /OD _{IL17A}]
	27-31-C04	286.79	703.62	7.93	21.31	0.41	0.49	1.19	19%

5

TABLE 4. Binding of rabbit monoclonal antibodies to IL-17F. The OD(450-690nm) for IL-17F binding by IgG (10 µg/ml) is calculated relative to a) OD(450-690nm) for secukinumab binding and b) OD(450-690nm) for IL-17A binding.

IgG	Relative binding IL-17F compared to secukinumab at 10 µg/ml	Relative binding IL-17F compared to IL-17A at 10 µg/ml
Clone ID	Binding IL-17F rel. to 10 µg/ml secukinumab (%)	OD _{IL-17F} /OD _{IL-17A} (%)
27-31-C04	19.7%	19.1%

1.3.6 Selectivity for IL-17A versus IL-17F

Selectivity of IgG binding to IL-17A over other IL-17 family members was determined to ensure target-specific binding. To determine the binding of anti-IL-17A IgG to IL-17F a direct ELISA was developed in which the plate is coated with IL-17A or IL-17F (Peprotech Cat. No. 200-25) and incubated with 10 µg/ml recombinant anti-IL-17A IgG (including secukinumab as control). The OD(450-690nm) for IL-17F binding is calculated relative to a) OD(450-690nm) for IL-17A binding and b) OD(450-690nm) for secukinumab binding. Data is summarized in TABLE 4.

Example 2: Humanization and Generation of scFv

2.1 Generation of Humanized scFv Antibodies

Based on data obtained during hit confirmation, clones 27-07-G02 and 27-31-C04 were selected for humanization. The humanization of the selected clones comprised the transfer of the rabbit CDRs onto an scFv acceptor framework of the V κ 1- λ cap/VH3 type as described in WO 2014/206561. In this process, the amino acid sequence of the six CDR regions was identified on the donor sequence (rabbit mAb) and grafted into the acceptor scaffold sequence, resulting in the constructs termed “CDR graft” (see (see SEQ ID NO: 24, A1; and SEQ ID NO: 61, PRO571, respectively). In addition, a second graft was designed for both clones 27-07-G02 and 27-31-C04 (SEQ ID NO: 25, A2; and SEQ ID NO: 62, PRO592, respectively), which included additional amino acids modifications from the rabbit donor in certain framework positions, which have been described to potentially influence CDR positioning and thus antigen binding and/or stability (Borras et al., 2010; J. Biol. Chem., 285:9054-9066). These humanized construct are termed “structural (STR) graft”. Further, in the case of clone 27-07-G02, four mutations, namely A51P on variable light chain, and Q14K, G16E, and G56A on variable heavy chain (according to AHo numbering) were introduced in the structure-based graft (SEQ ID NO: 25, A2) in order to improve affinity. In the case of clone 27-31-C04, two mutations, namely R20T and Q141P (according to AHo numbering) were introduced in variable heavy chain of the structure-based graft SEQ ID NO: 62, PRO592) in order to improve affinity.

In case the comparison of the characterization data for these constructs revealed a significant advantage of the STR constructs additional variants can be designed that combine the CDR grafted VL with STR grafted VH. This combination has been proven to be often sufficient to retain the activity of the STR graft (Borras et al. JBC. 2010;285:9054-9066) and would generally be preferred as fewer non-human alterations in the human acceptor scaffold reduce the risk for impaired stability and also the potential for immunogenicity.

2.2 Manufacture of Humanized scFv

Once the in-silico construct design was completed, the corresponding genes were synthesized and bacterial expression vectors were constructed. The sequence of the expression constructs was confirmed on the level of the DNA and the constructs were manufactured according to generic expression and purification protocols.

The heterologous expression of the proteins was performed in *E.coli* as insoluble inclusion bodies by induced overnight expression in small scale (55 mL). The inclusion bodies were isolated from the homogenized cell pellet by a centrifugation protocol that included several washing steps to remove cell debris and other host cell impurities. The purified inclusion bodies were solubilized in a denaturing buffer (100 mM Tris/HCl pH 8.0, 6 M Gdn-HCl, 2 mM EDTA) and the scFvs were refolded by a scalable refolding protocol that generated milligram amounts of natively folded, monomeric scFv. At this point a standardized protocol was employed to purify the scFvs. The product after refolding was captured by an affinity chromatography to yield the purified scFvs. In addition, melting temperatures of scFvs were determined by differential scanning fluorimetry (DSF) measurement in order to support the selection of molecules entering more extensive stability assessment after completion of pharmacodynamic characterization described in the following paragraphs. DSF measurement of selected molecules is shown in more detail under paragraph 2.4.2.

2.3 Functional characterization of humanized scFvs

In the following the humanized scFvs were characterized for the primary pharmacodynamics properties, using the same assay systems as described for the Hit

confirmation phase, with certain adaptations though to accommodate for the different format of the scFv molecules.

2.3.1 Affinity to human IL-17A and to cynomolgus monkey IL-17A

Affinity of humanized scFvs to human IL-17A as well as Cynomolgus monkey (Macaca
5 fascicularis) IL-17A was determined by SPR analysis on a T200 device (Biacore, GE
Healthcare). In this experiment, biotinylated human IL-17A was captured using the Biotin-
CAPture kit from Biacore ("capture setup"). After each analyte injection cycle the CAP
sensor chip was regenerated and new antigen was captured. The scFvs were injected as
analyte using a dose response multi-cycle kinetic assay with concentrations of the analyte
10 ranging from 0.35 to 90 nM diluted in running buffer. Obtained sensorgrams were fitted using
the 1:1 binding model. In addition, the scFvs were analyzed in an alternative SPR assay setup
("direct setup"). IL-17A was immobilized on a CM5 sensor chip (GE Healthcare) by amine-
coupling. Serial dilutions of scFvs ranging from 0.35 to 90 nM were injected over the
immobilized IL-17A. When using the direct setup affinities were 2- to more than 50-fold
15 higher for the scFv tested compared to the capture setup. The lower affinities using the
capture setup could be due to interference of biotin with binding of the scFvs.

Cross-reactivity to cynomolgus IL-17A was measured with the IL-17A produced by
Trenzyme in a similar assay as used to measure binding to human IL-17A.

The scFvs A1 (CDR) and A2 (STR) bound to human IL-17A with affinities of 2.5×10^{-10}
20 and 3×10^{-9} M, respectively (TABLE 5), as measured in the SPR capture setup. In the direct
SPR setup, the scFvs A1 (CDR) and A2 (STR) bound to human IL-17A with affinities of 1.4×10^{-10}
and 5.5×10^{-11} M, respectively (TABLE 5). The scFvs A1 (CDR) and A2 (STR) bound
to Cynomolgus IL-17A with affinities of 9×10^{-10} and 5×10^{-9} M, respectively (TABLE 5).

The scFvs PRO571 (CDR) and PRO592 (STR) bound to human IL-17A with affinities of
25 1.2×10^{-10} and 3.7×10^{-10} M, respectively (TABLE 5), as measured in the SPR capture setup.
In the direct SPR setup, the scFvs PRO571 (CDR) and PRO592 (STR) bound to human IL-
17A with affinities of 4×10^{-11} and 3.8×10^{-11} M, respectively (TABLE 5). The scFvs

PRO571 (CDR) and PRO592 (STR) bound to Cynomolgus IL-17A with affinities of 3.3×10^{-11} and 1.6×10^{-10} M, respectively (TABLE 5).

2.3.2 HT-29 assay (neutralization of human IL-17A)

The HT29 assay tested the ability of the humanized scFvs to inhibit IL-17A-induced
5 GRO- α secretion of the human colon carcinoma cell line HT-29. As described above in
sections 1.3.3 and 1.3.5 TNF α was added in addition to IL-17A to increase the sensitivity of
the assay. 50'000 HT-29 cells were plated in each well of a 96-well plate. In addition to the
serially diluted scFvs as well as the internal reference secukinumab, pre-dilutions of human
TNF α (50 pg/ml) and human IL-17A (1 ng/ml) were added to the HT-29 cells. After 24 h
10 incubation at 37°C and 5% CO₂, supernatants were collected and GRO- α (CXCL1
chemokine) secretion was quantified by ELISA.

Inhibition curves for the humanized scFvs A1, PRO571 and PRO592 are shown in FIG.
4. All potency data are summarized in TABLE 5 and TABLE 6. The relative IC₅₀ values
reported for each molecule were calibrated to the reference antibody secukinumab to allow for
15 direct comparison of IC₅₀ values from different assay plates. Relative IC₅₀ values were
calculated in mass units (ng/ml) of secukinumab and the scFvs.

The humanized scFvs derived from clone 27-07-G02, A1 and A2, as well as the
humanized scFvs derived from clone 27-31-C04, PRO571 and PRO592, inhibited IL-17A-
induced GRO- α secretion with lower IC₅₀ than secukinumab (see TABLE 5 and TABLE 6),
20 and showed a more than 100-fold higher potency compared to secukinumab (see TABLE 5
and TABLE 6).

TABLE 5. Pharmacodynamic properties of the humanized scFvs.

Protein number	scFv		Affinity to human IL-17A (Capture setup)			Affinity to human IL-17A (Direct setup)		
	Clone ID		k_a ($M^{-1} s^{-1}$)	k_d (s^{-1})	K_D (M)	k_a ($M^{-1} s^{-1}$)	k_d (s^{-1})	K_D (M)
A1	27-07-G02-sc01		4,55E+05	1,12E-04	2,47E-10	7,37E+05	1,02E-04	1,39E-10
A2	27-07-G02-sc02		2,27E+05	6,97E-04	3,07E-09	1,08E+06	5,92E-05	5,51E-11

Protein number	Affinity to cyno IL-17A					$K_D, \text{cyno}/K_D, \text{human}$	Neutralization of IL-17A in HT-29 assay	Blocking of IL-17A/IL-17RA interaction
	scFv		$k_a \text{ (M}^{-1} \text{s}^{-1}\text{)}$	$k_d \text{ (s}^{-1}\text{)}$	$K_D \text{ (M)}$			
	Clone ID							
							rel. IC ₅₀ *	rel. IC ₅₀ *
A1	27-07-G02-sc01	2,88E+05	2,61E-04	9,05E-10	3,66	173,3		19,3
A2	27-07-G02-sc02	1,67E+05	8,68E-04	5,19E-09	1,69	375,4		24,0

*: IC₅₀, secukinumab/IC₅₀, Test Sample

Protein number	scFv		Affinity to human IL17A (Capture setup)			Affinity to human IL17A (Direct setup)		
	Clone ID		k_a ($M^{-1} s^{-1}$)	k_d (s^{-1})	K_D (M)	k_a ($M^{-1} s^{-1}$)	k_d (s^{-1})	K_D (M)
PRO571	27-31-C04-sc01		1.21E+06	1.44E-04	1.19E-10	1.13E+06	4.55E-05	4.03E-11
PRO592	27-31-C04-sc02		3.87E+05	1.42E-04	3.67E-10	1.08E+06	4.07E-05	3.76E-11

Protein number						$K_{D, cyno}/K_{D, human}$	Neutralization of IL17A in	Blocking of IL17A-
	scFv		Affinity to cyno IL17A				HT-29 assay	IL17RA interaction
	Clone ID	k_a (M ⁻¹ s ⁻¹)	k_d (s ⁻¹)	K_D (M)	rel. IC ₅₀ *		rel. IC ₅₀ *	
PRO571	27-31-C04-sc01	1.17E+06	3.88E-05	3.31E-11	0.28	190.8	22.9	
PRO592	27-31-C04-sc02	3.36E+05	5.38E-05	1.6E-10	0.44	196.0	40.9	

*: IC₅₀, secukinumab/IC₅₀, Test Sample

TABLE 6. Potencies of anti-IL-17A scFv to inhibit IL-17A-induced GRO- α secretion of the human colon carcinoma cell line HT-29.

Protein ID	scFv		Potency in HT-29 assay	
	Clone ID	IC ₅₀ [ng/mL]	rel. IC ₅₀ *	
A1	27-07-G02-sc01	0.2	173.3	
A2	27-07-G02-sc02	0.1	375.4	
*: IC ₅₀ , secukinumab/IC ₅₀ , Test Sample				
Protein ID	scFv		Potency in HT-29 assay	
	Clone ID	IC ₅₀ [ng/mL]	rel. IC ₅₀ *	
PRO571	27-31-C04-sc01	0.1	190.8	
PRO592	27-31-C04-sc02	0.13	196.0	
mea				

2.3.3 Competitive ELISA (inhibition of hIL-17A binding to hIL-17-RA)

In addition to the HT-29 assay, the potency of each humanized scFv to inhibit the interaction between human IL-17A and human IL-17RA was assessed by ELISA with the same procedure as described in section 1.3.4. Similarly to the HT-29 assay, individual IC₅₀ values on each plate are calibrated against the IC₅₀ of the reference molecule secukinumab that is taken along on each plate and relative IC₅₀ (relative IC₅₀: IC₅₀, secukinumab/IC₅₀, test antibody) (TABLE 5 and TABLE 7). In this assay 6 ng/ml IL-17A is implemented which means that the assay can only resolve potencies between IgGs with K_D > 193 pM. Inhibition curves for A1, A2, PRO571 and PRO592 are shown in FIG. 5 and potencies are summarized in TABLE 5 and TABLE 7.

TABLE 7. Potencies of anti-IL-17A scFv to block of the human IL-17A/IL-17RA interaction (ELISA).

Protein ID	scFv	Potency in IL-17A/IL-17RA ELISA	
	Clone ID	IC ₅₀ [ng/mL]	rel. IC ₅₀ *
A1	27-07-G02-sc01	2.0	19.3
A2	27-07-G02-sc02	1.6	24.0

*: IC₅₀, secukinumab/IC₅₀, Test Sample

Protein ID	scFv	Potency in IL-17A/IL-17RA ELISA	
	Clone ID	IC ₅₀ [ng/mL]	rel. IC ₅₀ *
PRO571	27-31-C04-sc01	1.8	22.9
PRO592	27-31-C04-sc02	0.9	40.9

*: IC₅₀, secukinumab/IC₅₀, Test Sample

2.3.4 Selectivity against IL-17B, IL-17C, IL-17D, IL-17E and IL-17F (competition ELISA)

The relative potential of IL-17B to F, as compared to IL-17A to half-maximally inhibit IL-17A binding to each scFv was assessed by competition ELISA. The potential to inhibit the interaction of biotinylated IL-17A with scFvs by unlabeled IL-17B, IL-17C, IL-17D, IL-17E and IL-17F was analyzed by competition ELISA. For this purpose, the scFvs were coated on a 96-well ELISA plate. Binding of biotinylated IL-17A to the coated scFvs in presence of serially diluted IL-17A or IL-17B to IL-17F was detected using the biotin-binding streptavidin conjugated to HRP. For the dose-response curve with IL-17A data were analyzed using a four-parameter logistic curve fit, and the concentration of IL-17A required to block the interaction of biotinylated IL-17A with the coated scFv by 50% (EC_{50}) was calculated. IL-17B to IL-17F did not show any significant inhibition of the interaction between biotinylated TNF and scFvs (see FIG. 6). To quantify the relative potential of these IL-17 family members as compared to IL-17A to inhibit IL-17A binding to each scFv the EC_{50} to inhibit the interaction by IL-17 family members relative to IL-17A was calculated. Since no significant inhibition was observed when using IL-17 members at an approximately 100 to 40'000-fold higher concentration than the EC_{50} of IL-17A, the selectivity for binding to IL-17A over IL-17 family members was determined to be significantly higher than 100 to 40'000-fold.

As shown in FIG. 6 and summarized in TABLE 8 for A1, PRO571 and PRO592, the interaction between the scFvs A1, PRO571 and PRO592 with biotinylated IL-17A was blocked by unlabeled IL-17A, while IL-17B, IL-17C, IL-17D, IL-17E and IL-17F did not show any significant effect even at the highest concentration of cytokine tested (20 to 500 μ g/ml). Hence, A1, PRO571 and PRO592 bind specifically to IL-17A but not to its closest homologues, IL-17B to IL-17F. Therefore, off-target binding of the scFvs described herein appears highly unlikely.

2.4 Biophysical Characterization of the humanized scFvs

2.4.1 Manufacture of stability material

The scFvs were produced again using the same manufacture process as described above (see section 2.2) at higher scale (1.2L-2.4 L expression volume). Additionally, protein samples were concentrated to >10 mg/mL using centrifugal concentration tubes after purification. High scale manufacture of material for stability assessment is compiled in
5 TABLE 9. All molecules could be produced in sufficient amounts and purity (>95% monomeric).

The producibility of the scFv constructs was characterized by different reporting points (TABLE 9). The producibility criterion shall ensure that the selected scFv entity can be expressed, refolded and purified in sufficient amounts to support later development of the lead
10 molecule. The defined criteria were the expression yield of scFv per liter of fermentation broth, as assessed by SDS-PAGE, and the purification yield achieved in the generic lab-scale process, as assessed by measurement of the amount of purified protein by UV spectrometry, calculated back to 1 liter of refolding solution.

The expression titer was determined on the level of the crude E.coli lysate after the
15 harvest of the cells by centrifugation. During the harvest a small loss of cells was anticipated, however, this factor was neglected for the calculation of the expression yield in favor of a more conservative estimation of the productivity. For the quantification of the scFv product in the lysate coomassie stained reducing SDS-PAGE was chosen due to the high specificity of the method that allows discriminating the product from the host cell proteins in the sample.

20 A second criterion to assess the producibility was the purification yield of scFv calculated per liter of refolding solution. This parameter addresses the potential bottleneck in the anticipated manufacturing process that includes a protein refolding step. Since the efficiency of the refolding procedure has proven to be limiting in comparable manufacturing processes the performance of the different constructs can be compared with respect to the producibility
25 normalized to a defined refolding volume. For the calculation of the yield the final protein sample from each batch was quantified by UV absorbance and divided by the actual refolding volume of the respective purification.

TABLE 8. Target selectivity of the humanized scFvs.

Protein ID	scFv Clone ID	Selectivity IL-17B		Selectivity IL-17C		Selectivity IL-17D	
		rel. IC50, IL-17B/IC50, IL-17A]	highest conc. tested (ng/ml)	rel. IC50, IL-17C/IC50, IL-17A]	highest conc. tested (ng/ml)	rel. IC50, IL-17D/IC50, IL-17A]	highest conc. tested (ng/ml)
A1	27-07-G02-sc01	>> 1307	100000	>> 1307	100000	>> 11871	500000
PRO571	27-31-C04-sc01	>> 8636	100000	>> 8636	100000	>> 42808	500000
PRO592	27-31-C04-sc02	>> 1102	100000	>> 1102	100000	>> 7716	500000

Protein ID	scFv Clone ID	Selectivity IL-17E		Selectivity IL-17F	
		rel. IC50, IL-17E/IC50, IL-17A]	highest conc. tested (ng/ml)	rel. IC50, IL-17F/IC50, IL-17A]	highest conc. tested (ng/ml)
A1	27-07-G02-sc01	>> 475	20000	>> 475	20000
PRO571	27-31-C04-sc01	>> 1712	20000	>> 1712	20000
PRO592	27-31-C04-sc02	>> 309	20000	>> 309	20000

TABLE 8. Manufacture of material for stability assessment.

Protein ID	Construct ID	Expression volume [mL]	Yield post Capto L [mg]	Final yield [mg]	Yield per L refolding [mg/L]	Yield per L expression [mg/L]	Purity SE-HPLC [% monomer]
A1	27-07-G02-sc01	1200	126.8	26.5	13.3	22.1	99.9
PRO592	27-31-C04-sc02	1200	74.0	51.7	25.8	43.1	97.6
PRO571	27-31-C04-sc01	2400	124.0	88.5	22.1	36.9	98.9

2.4.2 Thermal unfolding

Thermal unfolding curves obtained from DSF measurements of the scFvs constructs A1, PRO571 and PRO592 are presented in FIG. 7. For DSF measurement material produced in small scale expression was used. Samples in 50mM phosphate-citrate buffer at pH6.4 were prepared at a final protein concentration of 50 µg/mL and a final concentration of 5x SYPRO® Orange in a total volume of 100 µl. Twenty-five microliters of prepared samples were added in triplicate to white-walled AB gene PCR plates. The assay was performed in a qPCR machine used as a thermal cycler, and the fluorescence emission was detected using the software's custom dye calibration routine. The PCR plate containing the test samples was subjected to a temperature ramp from 25°C to 96°C in increments of 1°C with 30 s pauses after each temperature increment. The total assay time was about two hours. The T_m was calculated by the software GraphPad Prism using a mathematical second derivative method to calculate the inflection point of the curve. Measurements were done in duplicates, or in triplicates, respectively, as shown by error bars. Resulting T_m values have been determined by fitting of data to a Boltzmann equation.

FIG. 7 shows melting curve of selected domain which was used for calculation of T_m by fitting the data to a Boltzmann equation. TABLE 10 summarizes calculated melting temperature and purities measured by SE-HPLC of A1, PRO571 and PRO592.

TABLE 9. Monomeric content and T_m of selected domains.

Protein ID	Purity SE-HPLC [% monomer]	T _m [°C]
A1	95.1	74.3
PRO592	99.7	66.1
PRO571	98.9	68.8

2.4.3 Storage stability study

The scFvs A1, PRO571 and PRO592 were subjected to further stability studies such as a four-week stability study, in which the scFv was be formulated in an aqueous buffer at 10 mg/ml and stored at < -65°C, 4°C and 37°C for four weeks. At one week, two weeks and at the end of each study, the fraction of monomers and oligomers in the formulation were

evaluated by integration of SE-HPLC peak areas. TABLE 11 compares endpoint measurements obtained at d28 of the study. Monomeric content in % and protein concentration in mg/mL over time course of 28 days are shown in FIG. 8. The scFvs of the invention were shown to have 5% or less monomer loss, and in particular in the case of A1 less than 1% monomer loss, upon storage at a temperature of 4°C and a concentration of >10 mg/ml. Furthermore, the scFv A1 of the invention was shown to have <5% monomer loss upon storage at a temperature of 37°C and a concentration of >10 mg/ml.

TABLE 10. 4w stability study at 10 mg/mL and storage temperatures of 37°C, 4°C and -80°C.

Protein ID	Temp. [°C]	Conc. [mg/mL]	[% monomer content]		% monomer loss	% content loss
			d0	d28	d28	d28
A1	37	>10		97.2	2.72	-3.51
	4		99.9	99.6	0.3	-1.76
	-80			99.8	0.09	4.16
592 PRO	37	>10		71.9	26.3	4.2
	4		97.6	93.4	4.26	5.03
	-80			95.1	2.53	2.38
571 PRO	37	>10		74.4	24.76	1.71
	4		98.9	95.1	3.84	2.52
	-80			96.8	2.09	0.49

2.4.4 Freeze-thaw stability study

In addition to the storage stability study described above the compatibility of the scFvs A1, PRO571 and PRO592 were assessed with respect to freeze-thawing (F/T) cycles (colloidal stability).

For the F/T stability assessment the same analytical methods and parameters (% monomer content and % monomer loss) as for the storage stability study (SE-HPLC, SDS-PAGE) were applied to monitor the quality of the molecules after five F/T cycles. Samples were formulated in PBS and were concentrated using VivaSpin concentration devices to 10 mg/mL prior to study initiation. Small sample volumes (<20µL) allowed rapid freezing and thawing intervals. Samples were subjected to five repeated freeze-thawing cycles. They were frozen at a temperature of -80°C and thawed at RT. To assess stability of A1, PRO571 and PRO592, after each cycle analytical read-outs, such as protein content and purity, were

recorded at different time points by SE-HPLC and UV₂₈₀ measurement. Data are presented in TABLE 12. FIG. 9 illustrates the course of monomer content in % over five repeated F/T cycles.

2.4.5 pH stability study

5 The scFvs A1, PRO571 and PRO592 were subjected to a short-term stress stability study, in which the scFv molecules were formulated at 20 mg/ml in a set of aqueous (phosphate-citrate) buffer systems with pH values between 3.5 and 7.5. Monomeric content in % and % monomer loss was analyzed after storage for 48h at 4°C and 8h at 25°C (TABLE 13TABLE 12). A1, PRO571 and PRO592 exhibited <5% monomer loss in any of the tested buffer
10 systems at any timepoint measured (TABLE 13 and TABLE 14).

It is important to note that the different studies suggested within the scope of this assessment address distinct mechanistic aspects of protein stability. While both methods are designed to give an estimation of the potential product shelf life and stability the mechanisms addressed are profoundly different. The midpoint of transition (T_m) assessed by thermal
15 unfolding is a qualitative measure for protein domain stability (does not allow for thermodynamic determination of ΔG). Highly stable protein domains (high T_m) are less likely to spontaneously unfold at ambient temperature and thus less prone to irreversible aggregation/precipitation driven by unfolded domain interactions. High domain stability indicates dense packaging of amino acid residues, which also correlates with resistance
20 towards protease cleavage. The SE-HPLC assessment on the other hand quantitatively determines the content of the monomeric fraction as well as of soluble oligomers/aggregates. Such soluble oligomers are oftentimes reversible and relatively loose associations driven by electrostatic or hydrophobic interactions between correctly folded proteins. There is some correlation between T_m as assessed by thermal unfolding and the propensity for
25 oligomer/aggregate formation as assessed by SE-HPLC particularly for proteins with “border line” stability. Beyond a certain threshold T_m of approximately 60°C antibody variable domains are generally sufficiently stable to be resistant toward aggregation/precipitation and proteolytic degradation due to partial domain unfolding at ambient temperature.

Oligomerization driven by hydrophobic and/or electrostatic interactions of surface residues may, however, still occur. Importantly, in an accelerated (stress) stability study at elevated temperature (e.g. 37°C) the various mechanisms of oligomer formation and precipitation may occur simultaneously.

TABLE 11. Freeze-thaw stability assessment of selected clones.

TABLE 11. Freeze-thaw stability assessment of selected clones.								
Protein ID	Freeze/ Thaw cycles	Conc. [mg/mL]	[% monomer content]					
			FT0	FT1	FT2	FT3	FT4	FT5
A1	5	10	99.9	99.9	99.9	99.9	99.9	99.9
PRO592	5	10	97.6	96.1	95.6	95.4	95.2	95.2
PRO571	5	10	98.9	98.8	98.8	98.6	98.5	98.5
Protein ID	Freeze/ Thaw cycles	Conc. [mg/mL]	% monomer loss					
			FT1	FT2	FT3	FT4	FT5	
A1	5	10	0.0	0.0	0.0	0.0	0.0	
PRO592	5	10	1.5	2.0	2.2	2.4	2.4	
PRO571	5	10	0.1	0.1	0.3	0.4	0.4	

TABLE 12. Monitoring of monomeric content and monomer loss at two defined time points (t8, t48) and temperatures (4°C, 25°C) during short-term pH stress stability.

Protein ID	pH	[% monomer content]			% monomer loss	
		t0	t8	t48	t8	t48
A1	3.5	99.7	99.6	99.6	0.2	0.2
	4.5	99.6	99.6	99.5	0.0	0.1
	5.5	99.5	99.5	99.4	0.0	0.1
	6.5	99.7	99.5	99.5	0.1	0.2
	7.5	99.6	99.5	99.4	0.1	0.2
PRO571	3.5	98.0	95.6	95.7	2.5	2.3
	4.5	97.9	97.1	97.0	0.8	0.9
	5.5	96.5	95.8	95.4	0.8	1.1
	6.5	97.4	95.6	95.0	1.9	2.5
	7.5	96.2	95.2	95.0	1.0	1.3
PRO592	3.5	99.5	98.3	98.4	1.2	1.1
	4.5	99.5	99.0	99.1	0.5	0.5
	5.5	99.5	98.8	98.8	0.7	0.7
	6.5	99.5	98.7	98.7	0.8	0.8
	7.5	99.5	98.8	98.8	0.7	0.7

TABLE 13. Monitoring of protein concentration at two defined time points (t8, t48) and temperatures (4°C, 25°C) during short-term pH stress stability

Protein ID	pH	protein concentration [mg/mL]			% content loss	
		t0	t8	t48	t8	t48
A1	3.5	99.7	99.6	99.6	0.2	0.2
	4.5	99.6	99.6	99.5	0.0	0.1
	5.5	99.5	99.5	99.4	0.0	0.1
	6.5	99.7	99.5	99.5	0.1	0.2
	7.5	99.6	99.5	99.4	0.1	0.2
PRO571	3.5	27.7	26.4	26.3	4.8	5.0
	4.5	26.5	27.1	26.2	-2.3	1.1
	5.5	29.3	27.9	28.4	4.8	3.1
	6.5	29.5	27.4	29.2	7.4	1.1
	7.5	29.3	29.3	29.2	0.1	0.2
PRO592	3.5	28.3	27.8	28.0	1.8	1.1
	4.5	26.2	25.1	26.4	4.5	-0.7
	5.5	25.3	25.8	25.3	-2.0	0.1
	6.5	28.4	27.7	28.1	2.5	1.1
	7.5	28.7	28.4	28.5	1.0	0.9

TABLE 14. Summary of the biophysical characterization data.

Protein ID	Clone ID	Tm [°C]	%	%	%	%	%	%
			monomer loss, 10 mg/mL, 5xFT	monomer loss, 28d, 10 mg/mL, 37°C	monomer loss, 28d, 10 mg/mL, 4°C	monomer loss, 28d, 10 mg/mL, -80°C	monomer loss, pH3.5-7.5, 20 mg/mL, RT, t8	monomer loss, pH3.5-7.5, 20 mg/mL, 4°C, t48
A1	27_07_G02_sc01	71.2	0.0	2.72	0.3	0.09	0.0-0.2	0.1-0.2
PRO592	27_31_C04_sc02	68.4	2.4	26.3	4.26	2.53	0.5-1.2	0.5-1.1
PRO571	27_31_C04_sc01	68.8	0.4	24.76	3.84	2.09	0.8-2.5	0.9-2.3

5 2.4.6 Methods

2.4.6.1 Reducing SDS-PAGE

Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis (SDS-PAGE) is an analysis technique used for qualitative characterization and to control purity of proteins. According to the United States Pharmacopeia (USP) (USP Chapter 1056) analytical gel electrophoresis is an appropriate and routine method to identify and to assess the homogeneity of proteins in drug substances.

The method is used to quantify the amount of scFv product from E.coli lysates to derive the expression yield after fermentation. Another application of the method is to verify the identity of test substances based on their molecular weight with respect to the theoretical values. For supportive purposes this method is used to quantify the purity of test samples with
5 respect to process-related impurities (host cell proteins) and product related impurities (degradation products or adducts).

The SDS-PAGE analyses were performed with commercially available precast gel system "Mini Protean" obtained from Bio-Rad Laboratories Inc. Humanized scFvs were analyzed on "Any kD" resolving gels (#456-9036). In both cases the Tris/Glycine buffer
10 system recommended by the manufacturer was used. For the detection of protein bands coomassie staining with SimplyBlue™ staining solution (Life Technologies Corp., #LC6060) was employed. For the staining procedures the protocols of the supplier was followed.

The documentation and analysis of the stained protein gels was performed with the
15 documentation system ChemiDoc XRS System (Bio-Rad Laboratories Inc., #170-8265) and software Image Lab, Version 4.0.1 (Bio-Rad Laboratories Inc., # 170-9690).

2.4.6.2 UV Absorbance at 280 nm

The method UV absorbance at 280 nm is a total protein assay as outlined in USP Chapter 1057. Protein solutions absorb UV light at a wavelength of 280 nm due to the presence of
20 aromatic amino acids. The UV absorbance is a function of the content of tyrosine and tryptophan residues in the protein and is proportional to the protein concentration. The absorbance of an unknown protein solution can be determined according to USP Chapter 851 on spectroscopy by applying Beer's law: $A = \epsilon \cdot l \cdot c$, where the absorbance (A) is equal to the product of the molar absorptivity (ϵ), the absorption path length and the concentration of the
25 substance. The molar absorptivity for the scFv was calculated with the software Vector NTI® (Life Technologies Corporation).

The measurement of the UV absorbance is performed with the Infinity reader M200 Pro equipped with Nanoquant plate (Tecan Group Ltd.). The absorbance of the protein samples is measured at 280 nm and 310 nm, where the latter wavelength is serving as a reference signal that is subtracted from the 280 nm signal. To account for potential interference of the sample matrix a blank subtraction is performed for each measurement. The final absorbance signal of a protein sample obtained is used to calculate the protein concentration using Lambert-Beer's law.

All measurements are performed within the range given by the instruments specifications in the measurement range of 0-4 OD, where a reproducibility of < 1% and a uniformity of < 3% is specified by the manufacturer.

2.4.6.3 SE-HPLC (Size Exclusion High-pressure Liquid Chromatography)

SE-HPLC is a separation technique based on a solid stationary phase and a liquid mobile phase as outlined by the USP chapter 621. This method separates molecules based on their size and shape utilizing a hydrophobic stationary phase and aqueous mobile phase. The separation of molecules is occurring between the void volume (V₀) and the total permeation volume (V_T) of a specific column. Measurements by SE-HPLC are performed on a Chromaster HPLC system (Hitachi High-Technologies Corporation) equipped with automated sample injection and a UV detector set to the detection wavelength of 280 nm. The equipment is controlled by the software EZChrom Elite (Agilent Technologies, Version 3.3.2 SP2) which also supports analysis of resulting chromatograms. Protein samples are cleared by centrifugation and kept at a temperature of 4-6°C in the autosampler prior to injection. For the analysis of scFv samples the column Shodex KW402.5-4F (Showa Denko Inc., #F6989201) is employed with a standardized buffered saline mobile phase (50 mM Sodium acetate pH 6.0, 250 mM sodium chloride) at the recommended flow rate of 0.35 mL/min. The target sample load per injection was 5 µg. Samples are detected by an UV detector at a wavelength of 280 nm and the data recorded by a suitable software suite. The resulting chromatograms are analyzed in the range of V₀ to V_T thereby excluding matrix associated peaks with >10 min elution time.

To ensure intermediate precision of the method, a reference standard is routinely measured at the beginning and end of each HPLC sequence. The reference standard used for this system suitability test is an scFv that has been produced as a batch and is aliquoted to be used for each measurement timepoint.

5 **2.4.6.4 DSF (Differential Scanning Fluorimetry)**

The midpoint of transition for the thermal unfolding of the tested scFv constructs was determined by Differential Scanning Fluorimetry (DSF), essentially as described by Niesen (Niesen et al., Nat Protoc. 2 (2007) 2212-21). The method DSF is a non-compensated method to measure temperature-dependent protein unfolding. The measurement of the thermal
10 unfolding temperature by DSF are performed with a MX3005P qPCR machine (Agilent Technologies) controlled with the MX Pro software package (Agilent Technologies) and equipped with an excitation/emission filter set at 492/610 nm. The reactions are set-up in Thermo fast 96 white PCR plates (Abgene; #AB-0600/W). For the detection of protein unfolding a commercially available stock solution of the dye SYPRO orange (Molecular
15 Probes; # S6650) is used at a final dilution of 1:1'000. The protein samples are diluted for the unfolding measurements to a final concentration of 50 µg/mL in a standardized buffered saline solution. The thermal unfolding is performed by a temperature program starting at 25°C ramping up to 96°C in 1°C steps with a duration of 30 seconds. During the temperature program the fluorescence emission of each sample is recorded. The recorded raw data is
20 processed and evaluated with a package of Microsoft Excel templates (Niesen, Nature Protocols 2007, Vol. 2 No.9) and the fluorescence data is fitted with a Boltzmann equation using the program GraphPad Prism (GraphPad Software, Inc.) to obtain the midpoint of transition (T_m).

In order to produce reliable and robust measurements of the midpoint of unfolding at
25 least duplicate measurements are performed. For an assessment of the intermediate precision a reference standard (known characterized scFv) is included with every measurement to allow for comparison of assay performance on different days.

2.4.6.5 Stability Study

In order to assess the stability of different scFv constructs as a read-out for the developability of these molecules a short-term stability study protocol can be designed. The protein constructs are concentrated in a simple buffered saline formulation (see above) to the target concentrations of 10 mg/mL. The monomer content is determined by SE-HPLC to confirm that the purity is exceeding the success criteria of > 95%. Subsequently the protein samples are stored at <-80°C, 4°C and 37°C for the duration of 4 weeks and aliquots are analyzed at various time points. The primary read-out is the analysis by SE-HPLC, which allows the quantification of soluble higher molecular weight oligomers and aggregates. As supportive measurements the protein content is determined by UV absorbance at 280 nm, which gives an indication whether during the storage period substantial amounts of protein were lost by precipitation. Additionally, purity at the end point of the study is determined by SDS-PAGE that indicates the stability of the construct with respect to degradation or covalent multimerization.

Example 3: Multispecific construct generation

Anti-IL-17 (27-07-G02-sc01, see TABLE 1), anti-TNF α (16-19-B11-sc06, see TABLE 1, described in WO 2017/158101) and anti-HSA-binding domains (19-01-H04-sc03, see see TABLE 1) were combined into two different trispecific antibody formats: Tribody and scDb-scFv formats (TABLE 1). The tribody format is a fusion of a Fab fragment with one scFv molecule fused to the carboxy terminus of each chain of the Fab resulting in a trispecific molecule (Fab-(scFv)₂). The format employs a heterodimeric assembly of a Fab fragment consisting of a variable light domain (VL) fused to a constant light domain (CL) on one protein chain and a variable heavy domain (VH) fused to a constant heavy domain (CH1) on the second protein chain. At the carboxy terminus, constant domains are each connected via a flexible Gly-Ser linker to an scFv fragment. The scDb-scFv is a format in which an scFv fragment is fused by a flexible Gly-Ser linker to the highly stable single chain diabody (scDb) format. Both formats, the Fab-scFv₂ (tribody) and the scDb-scFv can be expressed recombinantly in CHO-S host cells.

The optimal relative location of the individual binding domains in the trispecific formats was tested in terms of pharmacodynamic and biophysical parameters. As each format allows for different configurations, i.e. relative positioning of the domains to each other.

ScFv fusions at positions CL and CH1 of the tribody fragment are considered as equivalent, resulting in three variants of this format (FIG.10, A). Because the C-terminal fusions of the scFv moiety to the single chain Diabody (scDb) core domain of the molecule are superior to N-terminal fusions, only C-terminal fusion variants of this format were evaluated. Several permutation-variants of this format were analyzed (FIG. 10, B). Sequences of all molecules are listed in TABLE 1.

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Example 4: Lead manufacture generic process

Expression of all trispecific constructs was performed in CHO-S cells using CHOgro transient transfection kit (Mirus). Cultures were harvested after 5-7 days (cell viability <70%) of expression at 37°C by centrifugation and proteins were purified from clarified culture supernatants by Protein L affinity chromatography followed by a polishing step by size-exclusion chromatography. For the quality control of the manufactured material standard analytical methods, such as SE-HPLC, SDS-PAGE, and UV280 were used.

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TABLE 16 outlines production and characterization summary of trispecific format variants. FIG. 11 outlines production and purification of A3-A8. Antibody fragments were captured directly from CHO cell culture supernatant using a packed Protein L column pre-equilibrated in phosphate buffered saline (PBS) binding buffer. After sample application, column was washed with binding buffer, and protein was eluted with a one-step pH gradient. Monomeric content of protein fractions was determined by size-exclusion high-performance liquid chromatography (SE-HPLC) and fractions were pooled and buffer exchanged to phosphate citrate buffer at pH6.4 (Final Buffer) by dialysis. Fraction pools were concentrated using centrifugal concentrators to reduce total sample volume for subsequent preparative size exclusion chromatography (SEC) which was done for polishing of protein samples using a GE

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Superdex 200 10/300 column pre-equilibrated in PBS. Samples were eluted isocratically using Final Buffer and were further characterized by SE-HPLC, SDS-PAGE and UV₂₈₀.

TABLE 16. Production and characterization summary of trispecific format variants.

Protein ID	Format	Yield post ProteinL [mg/L culture]*	Purity SE-HPLC post SEC [% monomer]
A3	Tribody	65.9	98.4
A4	Tribody	17.5**	99.2
A5	Tribody	41.1	100.0
A6	scDb-scFv	36.3	98.9
A7	scDb-scFv	22.5	100.0
A8	scDb-scFv	29.6	98.9

*UV280

**~50% loss due to experimental error

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Example 5: Pharmacological Characterization

5.1 Affinity to HSA and CSA at pH 5.5 and pH 7.4

10 The binding to human serum albumin (HSA) and cynomolgus serum albumin (CSA) was assessed for purified trispecific molecules by running SPR spectroscopy on a MASS-1 device (Sierra Sensors). HSA was directly immobilized on a high capacity amine sensor chip (Sierra Sensors) and trispecific molecules were injected as analyte using a dose response multicycle kinetic assay with concentrations of the analyte ranging from 0.35 to 90 nM
15 diluted either in running buffer at pH5.5 or pH7.5. Obtained sensorgrams were fitted using the 1:1 binding model. Affinity to CSA was assessed in a similar approach using a CSA

immobilized sensor chip. Binding to CSA was measured for A5 and A7 at pH5.5 and pH7.5 only. The following table (TABLE 17) summarizes obtained affinities to HSA and CSA.

TABLE 17. Affinity to HSA and CSA at pH5.5 and pH7.4

Protein number	Affinity to HSA at pH5.5				Affinity to HSA at pH7.4			
	ka (M ⁻¹ s ⁻¹)	kd (s ⁻¹)	KD (M)	Binding (%)#	ka (M ⁻¹ s ⁻¹)	kd (s ⁻¹)	KD (M)	Binding (%)#
A3	6.23E+05	5.90E-04	9.47E-10	70.2%	4.06E+05	5.57E-04	1.372E-09	60.5%
A4	1.85E+05	3.08E-04	1.67E-09	51.5%	8.72E+04	4.17E-04	4.788E-09	43.3%
A5	1.80E+05	2.99E-04	1.66E-09	54.0%	1.10E+05	2.95E-04	2.686E-09	42.8%
A6	4.75E+05	3.65E-04	7.7E-10	62.3%	2.66E+05	3.32E-04	1.248E-09	57.5%
A7	2.84E+05	3.73E-04	1.32E-09	58.7%	1.93E+05	3.69E-04	1.913E-09	55.0%
A8	4.69E+05	3.75E-04	7.99E-10	62.2%	3.10E+05	4.40E-04	1.421E-09	56.3%

#: Maximum binding level achieved normalized to theoretical R_{max}

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Protein number	Affinity to CSA at pH5.5				Affinity to CSA at pH7.4			
	ka (M ⁻¹ s ⁻¹)	kd (s ⁻¹)	KD (M)	Binding (%)#	ka (M ⁻¹ s ⁻¹)	kd (s ⁻¹)	KD (M)	Binding (%)#
A3			not measured				not measured	
A4			not measured				not measured	
A5	1.81E+05	1.44E-04	7.952E-10	37.1%	7.78E+04	1.93E-04	2.474E-09	19.4%
A6			not measured				not measured	
A7	3.18E+05	1.47E-04	4.63E-10	44.1%	1.64E+05	2.15E-04	1.311E-09	31.6%
A8			not measured				not measured	

#: Maximum binding level achieved normalized to theoretical R_{max}

5.2 Affinity to human TNF α and to human and cynomolgus monkey IL-17A

The affinity to human TNF α and human and cynomolgus IL-17A was measured using SPR analysis on a T200 device (Biacore, GE Healthcare). In this experiment, the biotinylated antigens were captured using the Biotin-CAPture kit from Biacore. After each analyte injection cycle the CAP sensor chip was regenerated and new antigen was captured.

5 Trispecific molecules were injected as analyte using a dose response multicycle kinetic assay with concentrations of the analyte ranging from 0.35 to 90 nM diluted in running buffer. Obtained sensorgrams were fitted using the 1:1 binding model. Binding to cynomolgus IL-17A was measured only for A5 and A7. The following table (TABLE 18) summarizes affinities obtained for human TNF α and human and cynomolgus IL-17A. As shown in this
10 table, for some molecules binding to TNF α or IL-17A was lower than 10%. One can possibly explain this finding by reduced accessibility of the domains to their respective target. Although the measured affinity of these low binders was in a similar range and sometimes even significantly higher as compared to the molecules reaching higher binding levels the potential of the molecules to inhibit biological activity of TNF α and IL-17A was significantly
15 reduced (see TABLE 19 and TABLE 21).

The effect of the purified trispecific molecules on TNF-induced biological responses was determined using mouse L929 fibroblasts. First, L929 cells were growth-arrested with Actinomycin D. Then, 60'000 cells were plated per well and then treated with human (5 pM) or cynomolgus (5 pM) TNF α as well as with 3-fold serially diluted trispecific molecules and
20 the internal reference A13, an scDb containing the identical anti-TNF domain as the trispecific molecules (SEQ ID NO: 125). After 20h incubation cell viability was assessed using the cell counting kit-8 from Sigma-Aldrich. Data obtained for the neutralization of TNF α by the six trispecific molecules are depicted in FIG. 12. In addition, results obtained for A5 and A7 comparing their ability to neutralize human and cynomolgus TNF α are presented
25 in FIG. 13. Determined relative IC₅₀ values to the reference A13 for human TNF α as well as the ratio between the IC₅₀ values obtained for human and cyno TNF α are summarized in TABLE 19.

TABLE 18. Affinity to human TNF α and to human and cynomolgus IL-17A

Protein number	Affinity to human TNF α			Affinity to human IL-17A			Affinity to cynomolgous IL-17A					
	ka (M-1 s-1)	kd (s-1)	KD (M)	Binding (%)#	ka (M-1 s-1)	kd (s-1)	KD (M)	Binding (%)#	ka (M-1 s-1)	kd (s-1)	KD (M)	Binding (%)#
A3	4.31E+04	<1.00E-06	<2.32E-11	1.0%	5.91E+04	9.04E-05	1.53E-09	5.8%			not measured	
A4	9.08E+04	2.33E-05	2.57E-10	4.4%	2.15E+05	7.23E-05	3.37E-10	29.6%			not measured	
A5	2.91E+05	2.92E-05	1.00E-10	11.7%	1.39E+05	7.99E-05	5.75E-10	19.3%	7.39E+04	3.86E-05	5.23E-10	23.3%
A6	1.12E+05	2.66E-06	2.37E-11	6.0%	2.16E+05	8.96E-05	4.14E-10	30.2%			not measured	
A7	2.96E+05	3.59E-05	1.22E-10	14.0%	2.29E+05	7.83E-05	3.41E-10	31.6%	2.64E+05	5.89E-05	2.23E-10	51.5%
A8	2.19E+05	4.06E-05	1.85E-10	9.3%	1.73E+05	8.36E-05	4.83E-10	25.6%			not measured	
#: Maximum binding level achieved normalized to theoretical Rmax												

#: Maximum binding level achieved normalized to theoretical Rmax

TABLE 19. Summary table of relative potency data obtained in the L929 assay for the trispecific molecules.

Protein ID	Neutralization of TNF in L929 assay		
	Human		IC ₅₀ , cyno/IC ₅₀ , human
	IC ₅₀ [nM]	rel. IC ₅₀ & A13	
A3	4.5	0.003	n.a.
A4	0.08	0.2	n.a.
A5	0.02	0.9	3.3
A6	0.06	0.2	n.a.
A7	0.01	1	2.5
A8	0.03	0.4	n.a.

&: IC₅₀, A13/IC₅₀, Test Sample (nM)

5.3 HT-29 assay (concomitant blocking of human TNF and human IL-17A)

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IL-17A synergizes with TNF in up-regulating the expression of a subset of chemokines and cytokines such as Gro- α /CXCL1 in the colorectal adenocarcinoma cell line HT-29. Therefore, concomitant blockade of human TNF α and human IL-17A was assessed by analyzing Gro- α /CXCL1 secretion by HT-29 cells. To test the impact of HSA binding on

10 potency the IC₅₀ to inhibit Gro- α /CXCL1 secretion induced by IL-17A and TNF α was also assessed in presence of HSA. Experiments were conducted in absence and presence of 1 mg/ml HSA. 50'000 HT-29 cells were plated in each well of a 96-well plate. In addition to the titrated tested trispecific molecules as well as with the internal reference A13 (SEQ ID NO: 125) and secukinumab, pre-dilutions of human TNF α (0.2 ng/ml) and human IL-17A (2

15 ng/ml) were added to the HT-29 cells. After 24 h incubation at 37°C and 5% CO₂, supernatants were collected and GRO- α (CXCL1 chemokine) secretion was quantified by ELISA. The titration curves generated for the six trispecific molecules in presence of 1 mg/ml of human serum albumin (HSA) are presented in FIG. 14. Relative IC₅₀ values to both references secukinumab and A13 as well as the percentage of inhibition observed in presence

or absence of HSA are summarized in TABLE 20. Relative IC₅₀ to secukinumab was calculated by dividing the IC₅₀ of secukinumab in ng/ml by the IC₅₀ of the tested trispecific molecule in ng/ml. Relative IC₅₀ to A13 was calculated by dividing the IC₅₀ of A13 in nM by the IC₅₀ of the tested trispecific molecule in nM.

5 ***The best potency was observed for A5, A7 and A8. Potencies of trispecific molecules are 50 to 90-fold higher than secukinumab and 2 to 5-fold higher than A13. Concomitant blockade of TNFα and IL-17A drastically increased effect size compared to single blockade of TNFα or IL-17A. Approximately 200-fold and 60-fold lower concentrations are required to reach the same effect as the maximum effect achieved with secukinumab and A13,***
 10 ***respectively.***

TABLE20. Summary of relative potency data obtained in the HT-29 assay for the trispecific molecules.

Neutralization of IL-17A and TNF in HT-29 assay (-/+ HSA)						
Protein ID	rel. IC ₅₀ * secukinumab (ng/ml)		rel. IC ₅₀ & A13 (nM)		% Inhibition	
	-HSA	+HSA	-HSA	+HSA	-HSA	+HSA
A3	2.4	11.8	0.2	0.5	80.2	52.2
A4	7.1	6	0.6	0.3	106.6	104
A5	88.9	63	2.4	1.5	104.7	101.7
A6	30	26.8	0.6	1	105.1	98.9
A7	52	64.5	4.8	1	103.7	101.2
A8	44.1	304.2	3.9	4.6	103.4	100.4

*: IC₅₀, secukinumab/IC₅₀, Test Sample (ng/ml)

&: IC₅₀, A13/IC₅₀, Test Sample (nM)

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5.4 Competitive ELISA (inhibition of hIL-17A binding to hIL-17RA)

The inhibition of hIL-17A binding to hIL-17RA was assessed by competitive ELISA. hIL-17RA was coated on the ELISA plate. Biotinylated hIL-17A was preincubated with the trispecific molecules for 1.5 h, the mixture was then added to the ELISA plate to allow binding to the hIL-17RA for 1 h. Then, Streptavidin-HRP, used to detect the biotinylated IL-17A, was added and the plate was incubated for 1 h. Finally, Tetramethylbenzidine solution was added to develop the plate for 5 to 10 minutes and the reaction was stopped with 1 M HCl. The plate was read at 450 nm and 570 nm (reference wavelength). Titration curves obtained for the six trispecific molecules are represented in FIG. 15. Calculated relative IC₅₀ values to the reference secukinumab are summarized in TABLE 21.

TABLE 2115. Summary table of potency obtained in competition ELISA for trispecific molecules.

Protein number	Blocking of IL-17A/IL-17RA interaction
	rel. IC ₅₀ * secukinumab (ng/ml)
A3	2.2
A4	6.3
A5	4.9
A6	10
A7	10.6
A8	10.2

*: IC₅₀, secukinumab/IC₅₀, Test Sample (ng/ml)

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5.5 Simultaneous binding to TNF, IL-17A and HSA

To demonstrate simultaneous binding of the trispecific molecules to human TNF α , human IL-17A and HSA, a SPR experiment was employed. In brief, the six trispecific molecules (A3 – A8) were directly immobilized on a high capacity amine sensor chip (Sierra
5 Sensors) and the antigens were sequentially injected as analytes at concentrations of 2500 nM (HSA), 180 nM (TNF α) and 90 nM (IL-17A) ensuring that saturation of each binding was reached. All possible sequences of antigen injection were tested resulting in six possible sequences. For all trispecific molecules simultaneous binding to all three antigens was observed. Sensorgrams demonstrating the simultaneous binding to trispecific molecules are
10 shown in FIG. 16.

To evaluate concomitant binding of each antigen to trispecific molecules, the binding level relative to the theoretical R_{max} was calculated for each injection using the respective ligand and analyte molecular masses and the immobilization level of the trispecific molecules. The average of the maximum binding relative to theoretical R_{max} for each antigen was
15 obtained by averaging individual maximum binding relative to theoretical R_{max} obtained for the two injections where the respective antigen was injected as first analyte (in the absence of other bound antigens). Next, the binding relative to the theoretical R_{max} of each analyte injection was compared to its average maximum binding to calculate the fraction binding obtained in the presence of other bound antigens. The binding levels of each antigen relative
20 to its average maximum binding are shown in TABLE 12, which shows relative binding levels of each antigen compared to the average maximum binding. The binding levels relative to the theoretical R_{max} and the average of maximum binding (binding levels in the absence of other antigens) of each antigen were calculated. To evaluate binding of each antigen in the presence of other bound antigens, the binding relative to the theoretical R_{max} of each analyte
25 injection was normalized to its average maximum binding yielding a binding value that shows the degree of binding in the presence of other antigens.

TABLE 22. Relative binding levels of each antigen compared to the average maximum binding.

Binding levels of antigen relative to theoretical Rmax				Average of maximum binding levels relative to theoretical Rmax				Antigen binding levels obtained compared to average maximum binding levels relative to theoretical Rmax			
	Binding of first antigen	second antigen	Binding of third antigen					Binding of first antigen	Binding of second antigen	Binding of third antigen	
1. cycle	HSA	TNF α	IL-17A		HSA	TNF α	IL-17A		HSA	TNF α	IL-17A
A3	21%	34%	52%	A3	21%	37%	48%	A3	98,8%	92,8%	108,9%
A4	18%	37%	59%	A4	19%	39%	58%	A4	97,8%	94,2%	101,8%
A5	33%	43%	56%	A5	33%	44%	54%	A5	99,6%	97,8%	104,6%
A6	16%	21%	34%	A6	16%	22%	32%	A6	99,6%	94,1%	104,6%
A7	30%	23%	38%	A7	31%	25%	32%	A7	99,4%	92,6%	118,7%
A8	24%	24%	37%	A8	24%	26%	35%	A8	99,6%	93,7%	105,8%
2. cycle	HSA	IL-17A	TNF α						HSA	IL-17A	TNF α
A3	21%	43%	47%					A3	101,2%	89,3%	126,6%
A4	19%	53%	39%					A4	102,2%	91,9%	100,6%
A5	33%	50%	48%					A5	100,4%	92,9%	108,2%
A6	16%	29%	24%					A6	100,4%	91,3%	107,6%
A7	31%	28%	30%					A7	100,6%	85,4%	122,0%
A8	24%	31%	29%					A8	100,4%	88,9%	111,6%
3. cycle	TNF α	HSA	IL-17A						TNF α	HSA	IL-17A
A3	37%	21%	52%					A3	99,3%	98,7%	107,5%
A4	39%	14%	56%					A4	99,7%	77,7%	97,6%
A5	44%	30%	54%					A5	99,7%	92,7%	101,5%
A6	22%	14%	33%					A6	99,6%	84,5%	103,5%
A7	25%	26%	37%					A7	99,7%	86,1%	114,3%
A8	26%	21%	36%					A8	99,8%	84,9%	102,2%
4. cycle	TNF α	IL-17A	HSA						TNF α	IL-17A	HSA
A3	37%	53%	21%					A3	100,7%	110,8%	97,6%
A4	39%	58%	13%					A4	100,3%	100,5%	71,8%
A5	44%	56%	26%					A5	100,3%	105,5%	78,3%
A6	22%	35%	11%					A6	100,4%	107,8%	71,3%
A7	25%	39%	24%					A7	100,3%	122,5%	77,3%
A8	26%	39%	18%					A8	100,2%	110,1%	72,7%
5. cycle	IL-17A	HSA	TNF α						IL-17A	HSA	TNF α
A3	48%	21%	47%					A3	99,5%	100,8%	128,3%
A4	57%	17%	39%					A4	99,3%	91,5%	99,5%
A5	53%	28%	48%					A5	99,2%	85,0%	108,6%
A6	32%	13%	24%					A6	99,3%	83,1%	108,2%
A7	32%	28%	30%					A7	98,5%	90,5%	120,9%
A8	35%	21%	28%					A8	98,6%	88,2%	109,3%
6. cycle	IL-17A	TNF α	HSA						IL-17A	TNF α	HSA
A3	48%	47%	21%					A3	100,5%	128,8%	100,2%
A4	58%	39%	14%					A4	100,7%	99,6%	73,5%
A5	54%	48%	25%					A5	100,8%	108,2%	77,6%
A6	32%	24%	11%					A6	100,7%	110,3%	70,7%
A7	33%	30%	24%					A7	101,5%	122,1%	79,5%
A8	36%	29%	18%					A8	101,4%	113,6%	73,0%

5

For all combinations, the antigen binding was at least 70% of the average maximum binding relative to the theoretical Rmax (for example when TNF α was injected over

immobilized A3 as second or third analyte, the relative binding levels obtained were found either slightly decreased, to 92.8%, or slightly increased, to 126.6%, when compared to the binding levels in the absence of other antigens). Of note, when HSA was injected as the last analyte, the relative binding levels decreased to approximately 70% for all trispecific
5 molecules except for A3. In summary, simultaneous binding was observed for all trispecific molecules without substantial differences between the tested molecules.

5.6 Summary of pharmacokinetic characterization and molecule selection for basic developability assessment

10

TABLE 23 outlines pharmacological characterization of six trispecific molecules and provided basis for selection of molecules for developability assessment. Three molecules, namely A5, A7 and A8, were selected for developability assessment based on their overall better performance than A3, A4 and A6 in terms of TNF neutralization in L929 assay and
15 concomitant neutralization of TNF α and IL-17A in HT29 assay. All three molecules showed similar affinities to each of the three targets. A7 shows highest potency to neutralize both IL-17A and TNF α , while A5 shows approximately 2-fold reduced potency to neutralize IL-17A and A8 shows approximately 2-fold lower potency to neutralize TNF α compared to A7.

TABLE 23. Summary of pharmacological characterization.

Protein number	Affinity to human TNF		Affinity to human IL-17 (Capture setup)		Affinity to HSA at pH5.5		Neutralization of IL-17A and TNF in HT-29 assay						Blocking of IL-17A/IL-17RA interaction		Neutralization of TNF in L929 assay
	KD (M)	Binding (%)#	KD (M)	Binding (%)#	KD (M)	Binding (%)#	rel. IC50* secukinumab -HSA	+HSA	-HSA	+HSA	% Inhibition	rel. IC50* -HSA	+HSA		
A3	<2.32E-11	1.0%	1.53E-09	5.8%	9.467E-10	70.2%	2.4	11.8	0.2	0.5	80.2	52.2	2.2	0.003	
A4	2.57E-10	4.4%	3.37E-10	29.6%	1.67E-09	51.5%	7.1	6.0	0.6	0.3	106.6	104.0	6.3	0.2	
A5	1.00E-10	11.7%	5.75E-10	19.3%	1.659E-09	54.0%	88.9	63.0	2.4	1.5	104.7	101.7	4.9	0.9	
A6	2.37E-11	6.0%	4.14E-10	30.2%	7.697E-10	62.3%	30.0	26.8	0.6	1.0	105.1	98.9	10.0	0.2	
A7	1.22E-10	14.0%	3.41E-10	31.6%	1.316E-09	58.7%	52.0	64.5	4.8	1.0	103.7	101.2	10.6	1.0	
A8	1.85E-10	9.3%	4.83E-10	25.6%	7.991E-10	62.2%	44.1	304.2	3.9	4.6	103.4	100.4	10.2	0.4	

*: IC50, secukinumab/IC50, Test Sample (ng/ml)

&: IC50, A13/IC50, Test Sample (nM)

#: Maximum binding level achieved normalized to theoretical Rmax

Example 6: Developability Assessment

As A3, A4 and A6 displayed significantly reduced binding and potency of the anti-TNF domain, only A5, A7 and A8 were selected for developability assessment.

5

6.1 Exploratory pH compatibility

Selected purified trispecific candidates (A5, A7 and A8) were concentrated to >20 g/L and subsequently formulated in buffers covering the potential pH range of the formulation process. At these conditions ranging from pH 3.5 to 7.5, the molecules were incubated for either 8 h at 20°C or 48 h at 4°C before the generic stability indicating methods SE-HPLC and SDS-PAGE were applied to establish an initial compatibility profile of the molecules with potential process conditions and to define the buffer conditions for the storage stability assessment of the molecules.

The exploratory pH experiment was performed with A5 and A7 in multiple 50 mM phosphate-citrate buffers ranging from pH 3.5 to 7.5 (TABLE 24). In addition, the experiment was done in PBS buffer at pH7.4 with A5, A7 and A8 (TABLE 25). All three molecules did not lose >5% monomeric content in any of the buffers assessed.

TABLE 2416. Exploratory pH compatibility study in generic phosphate-citrate buffer.

Protein ID	pH	[% monomer content]			[% monomer loss]		Protein Concentration [mg/mL]			[% Content loss]	
		t0	t8	t48	t8	t48	t0	t8	t48	t8	t48
A5	pH 3.5	94.6	89.5	89.2	5.3	5.7	9.4	8.3	8.1	11.0	13.8
	pH 4.5	95.5	93.8	94.5	1.8	1.1	10.4	7.3	9.0	29.6	13.3
	pH 5.5	93.1	91.5	90.8	1.7	2.5	17.2	21.7	18.7	-25.9	-8.3
	pH 6.5	91.6	89.9	89.1	1.9	2.8	27.6	33.9	23.4	-22.7	15.5
	pH 7.5	90.8	87.7	88.2	3.4	2.9	43.9	22.8	53.3	48.1	-21.3
A7	pH 3.5	NA*	NA	NA	NA	NA	NA	NA	NA	NA	NA
	pH 4.5	98.6	98.0	97.8	0.6	0.8	0.8	-0.6	0.4	171.3	46.3
	pH 5.5	98.2	97.3	97.6	0.9	0.6	3.3	1.2	2.2	63.8	34.9
	pH 6.5	98.2	97.2	97.5	1.0	0.7	4.0	1.9	3.9	52.7	1.2
	pH 7.5	97.8	96.2	96.3	1.6	1.5	6.1	3.7	5.6	40.3	8.4

20

TABLE 25. Exploratory pH compatibility study in PBS.

Protein ID	[% monomer content]	[% monomer loss]
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	t0	t8	t48	t8	t48
A5	98.91	98.18	98.6	0.74	0.31
A7	97.67	94.62	92.81	3.12	4.98
A8	98.35	97.54	94.43	0.82	3.99

6.2 Thermal unfolding

Differential scanning fluorimetry (DSF) is a non-compensated method to measure temperature-dependent protein unfolding and was measured as described below.

Samples in 50mM phosphate-citrate buffer at pH6.4 were prepared at a final protein concentration of 50 µg/mL and a final concentration of 5x SYPRO® Orange in a total volume of 100 µl. Twenty-five microliters of prepared samples were added in triplicate to white-walled AB gene PCR plates. The assay was performed in a qPCR machine used as a thermal cycler, and the fluorescence emission was detected using the software's custom dye calibration routine. The PCR plate containing the test samples was subjected to a temperature ramp from 25°C to 96°C in increments of 1°C with 30 s pauses after each temperature increment. The total assay time was about two hours. The T_m was calculated by the software GraphPad Prism using a mathematical second derivative method to calculate the inflection point of the curve. The reported T_m is an average of three measurements.

TABLE 26 shows melting temperatures of trispecific molecules formulated in generic buffer (phosphate-citrate buffer at pH 6.4, 150 mM NaCl).

TABLE 26. DSF measurement of trispecific molecules in generic buffer.

Protein ID	Antibody Format	DSF in Generic buffer, T _m [°C]
A5	Tribody	64.3
A7	scDb-scFv	60.5
A8	scDb-scFv	59.0

20

6.3 Storage stability

As an indication for stability and ultimately developability of selected trispecific molecules a short-term stability study at different temperatures and protein concentrations in a generic non-optimized formulation buffer (PBS, pH7.4) was performed with selected molecules (A5, A7 and A8).

5 Samples were formulated in PBS and were concentrated using VivaSpin concentration devices to 10 mg/mL prior to study initiation. Incubation conditions applied for this study were temperatures of -80°C, 4°C and 37°C and protein concentrations of 10 mg/mL for a duration of 4 weeks. To assess stability of the molecules analytical read-outs such as protein content and purity were recorded at different time points by SE-HPLC, UV₂₈₀ measurement
10 and SDS-PAGE (FIG. 17 and TABLE 27).

TABLE 2717. Storage stability study performed at temperatures of 37°C, 4°C and -80°C at a protein concentration of 10 mg/mL for 4 weeks.

Protein ID	Temp. [°C]	Conc [mg/mL]	[% monomer content]						% monomer loss				
			d0	d2	d7	d14	d21	d28	d2	d7	d14	d21	d28
A5	-80.0	10.0	98.9	98.9	98.9	98.8	98.8	98.8	0.0	0.0	0.1	0.1	0.1
	4.0		98.9	98.9	98.7	98.2	98.0	97.8	0.0	0.2	0.7	0.9	1.1
	37.0		98.9	93.2	88.3	79.3	63.9	65.4	5.8	10.7	19.8	35.4	33.9
A7	-80.0	10.0	98.1	97.4	97.6	97.0	97.4	97.1	0.7	0.5	1.1	0.7	1.0
	4.0		98.1	96.2	95.2	94.6	94.6	94.1	1.9	3.0	3.6	3.6	4.1
	37.0		98.1	92.3	90.5	88.1	80.8	83.7	5.9	7.7	10.2	17.6	14.6
A8	-80.0	10.0	98.3	98.2	98.2	98.2	98.2	98.1	0.0	0.1	0.1	0.1	0.2
	4.0		98.3	98.0	97.2	95.9	95.3	95.0	0.3	1.1	2.4	3.0	3.3
	37.0		98.3	79.9	67.7	66.2	45.7	42.4	18.7	31.1	32.6	53.5	56.8

15

Recording of % monomeric content over time at 37°C revealed substantial differences between molecules as shown by overlays of SE-HPLC traces of d0 and d28 stability samples in FIG. 18. A8 was of clearly inferior stability, exhibiting a considerable amount high molecular weight species, when compared to A5 and A7, which is unexpected as the latter
20 molecule shares the same antibody format and consists of the same domains as A7 although with a permuted domain orientation.

6.4 Freeze / thaw stability

In addition to the storage stability study described above, the compatibility of the molecules was assessed with respect to five repeated freeze-thawing (F/T) cycles (TABLE 28). For the F/T stability assessment the same analysis methods as in the storage stability study were used to monitor the quality of the molecules after five F/T cycles. All molecules showed a loss in monomeric content below 1% (TABLE 28).

Samples were formulated in PBS and were concentrated using VivaSpin concentration devices to 10 mg/mL prior to study initiation. Small sample volumes (<20µL) allowed rapid freezing and thawing intervals. Samples were subjected to five repeated freeze-thawing cycles. They were frozen at a temperature of -80°C and thawed at RT. To assess stability of the molecules after each cycle analytical read-outs, such as protein content and purity, were recorded at different time points by SE-HPLC and UV₂₈₀ measurement.

TABLE 28. F/T stability assessment. Trispecific molecules were repeatedly frozen and thawed and monomeric content was measured after each cycle.

Protein ID	Temp. [°C]	Conc [mg/mL]	[% monomer content]					% monomer loss				
			FT1	FT2	FT3	FT4	FT5	FT1	FT2	FT3	FT4	FT5
A5	-80 / RT	10.0	98.9	98.9	98.8	98.8	98.8	0.0	0.0	0.1	0.1	0.1
A7	-80 / RT	10.0	97.4	97.6	97.0	97.4	97.1	0.7	0.5	1.1	0.7	1.0
A8	-80 / RT	10.0	98.2	98.2	98.2	98.2	98.1	0.0	0.1	0.1	0.1	0.2

6.5 Summary of basic developability assessment

Developability assessment is summarized in TABLE 29.

TABLE 29. Developability assessment.

Protein number	T _m	monomer loss 4w, 10 mg/mL @ 4 °C	monomer loss after 5 F/T cycles	monomer loss 8 h, ≥ 20 mg/mL @ 20 °C in any buffer system in pH range 3.5-7.5	monomer loss 48 h, ≥ 20 mg/mL @ 4 °C in any buffer system in pH range 3.5-7.5
A5	64.0°C	1.10%	0.10%	0.70%	0.30%
A7	60.5°C	4.10%	1.00%	3.10%	5.00%
A8	59.0°C	3.30%	0.20%	0.82%	3.99%
	> 60 °C	≤ 5%	≤ 5%	≤ 5%	≤ 5%

Example 7: Pharmacokinetic (PK) characterization in cynomolgus monkeys

To confirm prolongation of half-life with the use of a trispecific format comprising an anti-HSA domain, a non-GLP pharmacokinetic study in cynomolgus monkeys was performed. A7 was administered to cynomolgus monkeys according to the study plan briefly outlined below (see also TABLE 30). The pharmacokinetics of A7 were determined following intravenous and subcutaneous administration to male cynomolgus monkeys. A total of six non-naïve animals (three in each group) received a single administration of A7 at a target dose level of 3 mg/kg. The protein was pre-formulated in sterile 1x phosphate buffered saline, 150 mM L-Arginine, 500 mM sucrose, at pH 7.5 (TABLE 30).

TABLE 30. Overview of dosing regimen.

Group	Animal ID	Test Item	Route	Dose Level (mg/kg)	Dose Vol. (mL/kg)	Dose Conc. (mg/mL)
1	001M-003M	A7	IV	3	1.003	2.99
2	004M-006M	A7	SC	3	1.003	2.99

Serum was prepared from blood samples collected at the following timepoints: predose, 10 and 30 min and 1, 2, 4, 6, 8, 12, 24, 36, 48, 72, 96, 144, 192, 240, 288, 336, 384, 432 and 504 h post dose.

Measured serum concentrations are shown in FIG. 19. A strong decrease of A7 concentrations was observed from 192 hours on. All samples from timepoints 288 to 504h were below the limit of quantitation (BLQ). This decrease in signal suggested the development of anti-drug antibodies (ADA) by the monkeys. Formation of ADAs against human variable domains is expected and has been observed before with the anti-TNF α x HSA interim antibodies. Therefore, an assay was performed where A7 was coated on an ELISA plate and incubated with the serum samples. Cynomolgus IgGs bound to A7 were detected using an anti-monkey IgG antibody. The presence of ADA was confirmed and results are shown in FIG. 19. Since the animals developed ADAs from 192h on, later timepoints were omitted in the estimation of PD parameters in order to not underestimate the half-life of A7.

Monkey serum samples were subsequently analyzed in a PK-ELISA. Serum samples from all treated animals were diluted. ELISA plates were coated overnight with human TNF α and serial dilutions of A7 in 5% cynomolgus serum were added to the plates to yield a calibration curve. TNF α -bound A7 was detected with biotinylated human IL-17A followed by streptavidin poly-HRP. Serum samples with unknown concentrations were diluted 20-fold or more if required and A7 concentrations were interpolated from the calibration curve. For assay qualification, lower and upper limits of quantitation and quality controls were set to define an analytical range of 50 - 600 ng/ml. The assay was tested for dilution linearity, hook effect and selectivity. Additionally, stability of A7 in cynomolgus serum subjected to 1-3 freeze-thaw cycles was shown prior to quantification.

The data from PK analysis was then utilized for calculation of PK parameters. Pharmacokinetic parameters were estimated using WinNonlin pharmacokinetic software (Phoenix version 1.4) using a non-compartmental approach.

Accurate evaluation of different kinetic parameters requires the AUC from time zero to infinity. The extrapolation from the time of the last measured concentration (t_{last}) is done by mathematical integration of the curve from t_{last} to infinity. The extrapolated area from the last sample to infinity should be less than 20 % of the total AUC to confirm the accuracy of the derived kinetic parameters. For both IV and SC administration of A7, the extrapolation of the AUC to infinity represents more than 20% of the total area. As a result, the calculated data (TABLE 31) needs to be interpreted with caution. Most likely this effect arises due to the development of anti-drug antibodies, which interfere with the assay. Such interference could potentially be avoided through higher dosing.

TABLE 31. Pharmacokinetic parameters of A7 following intravenous and subcutaneous injection at a target dose level of 3 mg/kg.

Route (Target Dose)	Animal No.	C0 (ng/mL)	Tmax (h)	Cmax (ng/mL)	AUC(0-t) (h*ng/mL)	AUC(0- inf)A (h*ng/mL)	CL _A /CL _{FA} (mL/h/kg)	Vd _A / Vd _{FA} (mL/kg)	T1/2 _A (h)
IV (3 mg/kg)	001M	47000	4	63900	4920000	10000000	0.345	76.1	153
	002M	57400	0.5	76800	3570000	7740000	0.423	108	178
	003M	48300	2	53200	3750000	7230000	0.472	87.2	128
	Mean*	50900	2	64600	4080000	8330000	0.413	90.6	153
	SD	5680	-	11800	732000	1490000	0.0638	16.3	24.7
SC (3 mg/kg)	004M	-	36	43500	4390000	11100000	0.301	73.3	169
	005M	-	48	27800	3110000	5490000	0.603	93.6	108
	006M	-	24	33100	3420000	19100000	0.170	107	436
	Mean*	-	36	34800	3640000	11900000	0.358	91.2	237
	SD	-	-	8000	666000	6830000	0.222	16.9	175

* Median Tmax reported.

A Extrapolation of the AUC to infinity represents more than 20% of the total area. For 002M and 006M, the coefficient of determination of the elimination phase was <0.800. Values considered to be representative as detailed above.

NA Not applicable.

5

A clear half-life extension was observed for the scDb-scFv comprising an anti-HSA domain. Half-life ($t_{1/2}$) of A7 was more than 5 days. This was achieved independently of the mode of administration, as shown in TABLE 31. Due to stronger variations in the last points of the elimination phase (prior to signal drop), the half-life of 436 hours calculated for animal 10 006M might be less accurate than the half-lives determined for the remaining animals. Mean half-lives were higher after SC injection than after IV administration, with average half-lives of 237 hours (9.9 days) and of 153 hours (6.4 days) respectively.

Example 8: Complex formation analysis

15 In this study, various ratios of the scDb-scFv A7, and Morrison L A14 and A15 (see FIG. 20) and the target binding molecules, human IL-17A and human TNF α , were co-incubated and the generated complexes characterized.

A14 is a Morrison L format with the anti TNF α binding domains placed on the Fab arms and the anti IL-17A binding domains attached C-terminally to the light chains as scFvs.

20 A15 is a Morrison L format with the anti IL-17A binding domains placed on the Fab arms and the anti TNF α binding domains attached C-terminally to the light chains as scFvs.

Expression of A7, A14 and A15 was performed in FreeStyle CHO-S cells using the transient CHOgro expression system (Mirus). The genes of interest were optimized for mammalian expression, synthesized and cloned into a standard pcDNA3.1 vector. The signal 25 sequences originate from mouse heavy chain IgG. Expression cultures were cultivated in batch for 6 to 7 days (cell viability < 70%) either at 37 °C or for one day at 37 °C followed by a temperature shift to 32 °C for five to six days. The culture supernatant was separated by centrifugation followed by 0.45 μ m filtration. A7 was captured from the clarified culture supernatants by Protein L affinity chromatography followed by polishing size-exclusion 30 chromatography.

Recombinant human IL-17A was purchased from Peprotech, Rocky Hill, NJ, USA, as disulfide-linked homodimer expressed in *E. coli* and purified to a purity of $\geq 98\%$ by SDS-PAGE and HPLC (Catalog# 200-17, Lot# 061184).

Recombinant human TNF α was purchased from Peprotech, Rocky Hill, NJ, USA, as homotrimer expressed in *E. coli* and purified to a purity of $\geq 98\%$ by SDS-PAGE and HPLC (Catalog# 300-01A, Lot# 0906CY25).

The characterization of the formed complexes was conducted post-centrifugation with hydrodynamic radius determination by dynamic light scattering (DLS) and with protein concentration recovery by absorption at 280 nm measurement.

Dynamic Light Scattering is a fast and sensitive method to determine the size distribution of particles in solution by recording scattered light intensity fluctuations over time. The data is mathematically transformed to its autocorrelation function that yields the translational diffusion coefficient (D_t). The hydrodynamic radius (R_h) is subsequently derived from the Stokes-Einstein equation,

$$R_h = \frac{k_b T}{6\pi\eta D_t}$$

where k_b is the Boltzmann constant, T is the Temperature in Kelvin and η is the solvent viscosity.

The hydrodynamic radius was determined using the DynaPro® Plate Reader™ II and the Dynamics software package (Wyatt Technology Corp., Santa Barbara, CA, USA). 25 μ L were applied to a 384 well optical clear bottom microtiter plate (e.g. Corning® 384 well microplate, Merck, CLS3540-50EA) and remaining air bubbles removed by centrifugation (1000 g, 4 min). The samples were measured at 25 °C for 10 acquisitions of 5 to 10 s. The viscosity was set to PBS.

During the complex formation experiment, A7 was incubated with each of its soluble targets, human IL-17A and human TNF α , at the relative molar ratio given in TABLE 32 (relative numbers of binding sites (paratopes) on A7, and epitopes on the respective target), and a concentration at least 1000 fold above K_D (TABLE 32). The concentration range of each epitope/binding site and A7 was between 0.5 and 5 μ M.

TABLE 32: Relative number of paratopes and epitope in solution (Molar ratios for each target epitope and paratopes on A7)

Experiment	Molar ratios for paratope and epitopes		
	Antibody	human IL-17A	human TNF α
A	1	1	0
B	1	1	0.2
C	1	1	1
D	0.2	0.2	1
E	1	0	1
F	1	0.2	1
G	0.2	1	0.2
H	1	0.2	0.2
I	1	0	0

The hydrodynamic radii of the formed soluble complexes at each of the tested binding partner ratios are summarized in FIG. 21, FIG. 22 and FIG. 23. The percentage of recovered, soluble protein concentration is summarized in FIG. 24, FIG. 25 and FIG. 26.

1. For most of the conditions a complete recovery of protein concentration was observed that indicate no or limited formation of insoluble complexes for A7
2. Limited increase of hydrodynamic radius during complex size formation to maximum 5 fold for A7
3. Limited immunogenic risk because of soluble complexes formation and minor size increase for A7
4. Monovalent binding for each target by A7 facilitates the formation of only soluble complexes with minor size increase upon complex formation

Conclusions:

Antibody mediated immune complex formation has been associated with immunogenicity and immune related adverse events. Such immune complexes for example form, if a bivalent antibody cross-links a target molecule that exists in a multimeric form (i.e.

dimeric, trimeric, tetrameric, etc). In the situation of a trimeric target (e.g. TNF α) an antibody can bind with its two Fab arms to two different TNF trimers resulting in a small soluble complex. The two unbound units of each of the two engaged TNF trimers in the complex can in turn be bound by other antibodies. Upon concomitant engagement by multiple antibodies, 5 this leads to the formation of higher order complexes and immune precipitates. Such large immune complexes enhance immunogenicity of the therapeutic antibodies and can trigger the formation of anti-drug antibodies and immune related adverse events. The risk for immune complex formation becomes even more evident in the case where a molecule can simultaneously engage more than one target type, each by bivalent interactions. Examples for 10 this are the bispecific anti-TNF α /IL-17 blockers ABT122 and COVA322, which are interacting with each of their targets in a bivalent binding mode. While ABT122 was reported to trigger anti-drug antibody formation with high incidence, COVA322 even resulted in immune related adverse reactions (skin rashes), presumably triggered by immune complex formation.

15 Here we show that the bispecific monovalent scDb-scFv exemplified by A7 has a remarkably reduced capacity to mediate immune complex formation when compared to bispecific formats that interact in a bivalent manner with their targets (e.g. A14 and A15) comprising the same binding domains as A7. Thus we conclude that a monovalent bispecific format is preferred over a bispecific bivalent format due to its reduced risk for ADA 20 formation and its favorable risk-to-benefit profile.

Oftentimes, bivalent formats are chosen to compensate for the relatively low affinities to the target (e.g. COVA322 binding to IL-17). Therefore, in order to enable monovalent formats in multispecific approaches very high affinities to the respective targets are required. The high affinities of the Fv domains in A7 (and variants thereof) enable the use of a 25 monovalent format, resulting in a highly potent bi-specific therapeutic.

Further, the absence of an Fc region in the monovalent bispecific scDb-scFv format avoids binding to Fc receptors on immune cells, which may also contribute to the generation of immune related adverse events.

CLAIMS

1. An isolated multispecific antibody comprising a first domain specifically binding IL-17A, a second domain specifically binding TNF α , and, optionally, a third domain specifically binding to human serum albumin (HSA), wherein
5 (a) said first domain comprises (i) a set of CDRs: HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively, or (ii) a set of CDRs: HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 39, 40, and 41,
10 respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 50, 51, and 52, respectively; and
(b) said second domain comprises a set of CDRs: HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 63, 64, and 65, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 76, 77, and 78, respectively; and
15 (c) said third domain, when present, comprises a set of CDRs: (i) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 90, 91, and 92, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 100, 101, and 102, respectively; or
(ii) HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 111, 112, and 113, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 121, 122,
20 and 123, respectively.
2. The multispecific antibody of claim 1, wherein said antibody comprises only one domain specifically binding IL-17A and/or only one domain specifically binding TNF α , and/or, optionally, comprises only one domain specifically binding human
25 serum albumin.
3. The multispecific antibody of claim 1 or claim 2, wherein said domains are capable of binding to their respective antigen or receptor simultaneously.
- 30 4. The multispecific antibody of any one of the preceding claims, wherein said antibody has the following properties

- 5 (a) has the ability to neutralize IL-17A with a potency relative to that of secukinumab (relative potency), determined by measuring Gro- α secretion in an HT-29 assay, greater than 2, e.g. greater than 5, greater than 10, greater than 15, greater than 20, greater than 25, greater than 30, greater than 35, greater than 40, greater than 45, preferably greater than 50, and wherein said relative potency is the ratio of the IC₅₀ value in ng/mL of secukinumab as measured in the HT-29 assay to the IC₅₀ value in ng/mL of said multispecific antibody as measured in the HT-29 assay; and
- 10 (b) has the ability to neutralize TNF α with a potency relative to that of an scDb according to SEQ ID NO: 149 (A13) (relative potency), determined by measuring Gro- α secretion in an HT-29 assay, of at least 1, e.g., greater than 1, greater than 1.5, greater than 2, greater than 2.5, greater than 3, greater than 3.5, preferably greater than 4, more preferably greater than 4.5, and wherein said relative potency is the ratio of the IC₅₀ value in nM of said scDb according to SEQ ID NO: 149 as measured in the HT-29 assay to the IC₅₀ value in nM of said multispecific antibody as measured in the HT-29 assay; and
- 15 (c) optionally, has the ability to block interaction between IL-17A and IL-17RA with a potency relative to that of secukinumab (relative potency), determined in ELISA assay, greater than 2, e.g., greater than 3, greater than 4, greater than 5, greater than 6, greater than 7, greater than 8, greater than 9, preferably greater than 10, and
- 20 wherein said relative potency is the ratio of the IC₅₀ value in ng/mL of secukinumab as measured by ELISA to the IC₅₀ value in ng/mL of said multispecific antibody as measured by ELISA; and
- 25 (d) optionally, has the ability to neutralize TNF α with a potency relative to that of an scDb according to SEQ ID NO: 149 (A13) (relative potency), determined in L929 assay, of at least 0.4, e.g., at least 0.5, preferably at least 1, and wherein said relative potency is the ratio of the IC₅₀ value in nM of said scDb according to SEQ ID NO: 125 as measured in the L929 assay to the IC₅₀ value in nM of said multispecific antibody as measured in the L929 assay; and/or
- 30 (e) binds to human IL-17A with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, less than 2 nM, than 1 nM, preferably less than 0.5 nM, as measured by surface plasmon resonance; and optionally, binds to cynomolgus IL-17A with a K_D of less than 5 nM, e.g., less than 4 nM, less than 3 nM, less than 2 nM, than 1 nM, preferably less than 0.5 nM as measured by surface plasmon resonance;

- (f) binds to human TNF α with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, less than 2 nM, less than 1 nM, preferably less than 0.5 nM, more preferably less than 0.25 nM, as measured by surface plasmon resonance; and
- 5 (g) optionally, binds to human serum albumin with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, preferably less than 2 nM, as measured by surface plasmon resonance, and optionally, binds to cynomolgus serum albumin with a dissociation constant (K_D) of less than 5 nM, e.g., less than 4 nM, less than 3 nM, preferably less than 2 nM as measured by surface plasmon
- 10 resonance.
5. The multispecific antibody of any one of the preceding claims, wherein said antibody has the following properties
- (a) has a melting temperature (T_m), determined by differential scanning fluorimetry,
- 15 of at least 55°C, preferably of at least 58°C, more preferably at least 60°C in phosphate-citrate buffer at pH 6.4, 150 mM NaCl;
- (b) has a loss in monomer content, after five consecutive freeze-thaw cycles, of less than 5%, e.g., less than 4%, less than 3%, less than 2%, preferably 1% or less, when said multispecific antibody is at a starting concentration of 10 mg/ml in
- 20 phosphate buffered saline (PBS), pH 7.4;
- (c) has a loss in monomer content, after storage for at least two weeks, particularly for at least four weeks, at 4°C, of less than 10%, preferably less than 5%, when said multispecific antibody is at a starting concentration of 10 mg/ml in phosphate buffered saline (PBS), pH 7.4; and/or
- 25 (d) has a loss in monomer content, after storage for at least two weeks, particularly for at least four weeks, at 37°C, of less than 20%, preferably less than 15%, when said multispecific antibody is at a starting concentration of 10 mg/ml in phosphate buffered saline (PBS), pH 7.4.
- 30 6. The multispecific antibody of any one of the preceding claims, wherein said first domain and/or said second domain, and/or, optionally said third domain, comprise(s) a heavy chain variable region VH and wherein said VH is VH3 or VH4, preferably VH3.

7. The multispecific antibody of any one of the preceding claims, wherein said first domain and/or said second domain, and/or, optionally said third domain, comprise(s) a light chain variable region VL and wherein said VL comprises V κ frameworks FR1, FR2 and FR3, particularly V κ 1 or V κ 3 FR1 to FR3, preferably V κ 1 FR1 to FR3, and a framework FR4, which is selected from a V κ FR4, particularly V κ 1 FR4, V κ 3 FR4, and V λ FR4, particularly V λ FR4 comprising the amino acid sequence having at least 60, 70, 80, 90 percent identity to an amino acid sequence selected from any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in any of SEQ ID NO: 26 to SEQ ID NO: 32, preferably V λ FR4 as set forth in SEQ ID NO: 26 or 27, more preferably V λ FR4 as set forth in SEQ ID NO: 27.
8. The multispecific antibody of any one of claims 6 to 7, wherein said VH of the first domain comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 10; and/or said VL of the first domain comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 21, preferably wherein said VH of the first domain comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 10 and 11; and/or said VL of the first domain comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 21 and 22, more preferably wherein said VH of the first domain is as set forth in SEQ ID NO: 10 and/or said VL of the first domain is as set forth in of SEQ ID NO: 21.
9. The multispecific antibody of any one of claims 6 to 8, wherein said VH of the second domain comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence selected from the group consisting of SEQ ID NOs: 72, 73, 74 or 75, preferably SEQ ID NO: 72; and/or said VL of the second domain comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence selected from the group consisting of SEQ ID NOs: 85, 86, 87, 88 and 89, preferably SEQ ID NO: 85, preferably wherein said VH of the second domain comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 72, 73, 74 or 75; and/or said VL of the second domain comprises an amino acid

sequence selected from the group consisting of SEQ ID NOs: 85, 86, 87, 88 and 89, more preferably wherein said VH of the second domain is as set forth in SEQ ID NO: 72 and/or said VL of the second domain is as set forth SEQ ID NO: 85.

- 5 10. The multispecific antibody of any one of claims 6 to 9, wherein the third domain is present and
- (i) said VH of the third domain comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 99; and/or said VL of the
- 10 the amino acid sequence SEQ ID NO: 108, preferably wherein said VH of the third domain is as set forth in sequence of SEQ ID NO: 99 and/or a VL of the third domain is as set forth in SEQ ID NO: 108; or
- (ii) said VH of the third domain comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 120; and/or said VL
- 15 comprises an amino acid sequence that is at least 90 percent identical to the amino acid sequence SEQ ID NO: 130, preferably wherein said VH of the third domain is as set forth in SEQ ID NO: 120 and/or said VL of the third domain is as set forth in SEQ ID NO: 130.
- 20 11. The multispecific antibody of any one of the preceding claims, wherein said multispecific antibody is in a format selected from the group consisting of a single-chain diabody (scDb), a tandem scDb (Tandab), a linear dimeric scDb (LD-scDb), a circular dimeric scDb (CD-scDb), a bispecific T-cell engager (BiTE; tandem di-scFv), a tandem tri-scFv, a tribody (Fab-(scFv)₂) or bibody (Fab-(scFv)₁), Fab, , Fab-Fv₂,
- 25 Morrison (IgG CH3-scFv fusion (Morrison L) or IgG CL-scFv fusion (Morrison H)), triabody, scDb-scFv, bispecific Fab₂, di-miniantibody, tetraabody, scFv-Fc-scFv fusion, scFv-HSA-scFv fusion, di-diabody, DVD-Ig, COVD, IgG-scFab, scFab-dsscFv, Fv₂-Fc, IgG-scFv fusions, such as bsAb (scFv linked to C-terminus of light chain), Bs1Ab (scFv linked to N-terminus of light chain), Bs2Ab (scFv linked to N-terminus of heavy chain), Bs3Ab (scFv linked to C-terminus of heavy chain), Ts1Ab (scFv linked to N-
- 30 terminus of both heavy chain and light chain), Ts2Ab (dsscFv linked to C-terminus of heavy chain), Bispecific antibodies based on heterodimeric Fc domains, such as Knob-into-Hole antibodies (KiHs); an Fv, scFv, scDb, tandem-di-scFv, tandem tri-scFv, Fab-(scFv)₂, Fab-(scFv)₁, Fab, Fab-Fv₂, COVD fused to the N- and/or the C-terminus

of either chain of a heterodimeric Fc domain or any other heterodimerization domain, a MATCH and DuoBodies, preferably is tribody or scDb-scFv.

- 5 12. The multispecific antibody of any of the preceding claims, wherein the antibody comprises an amino acid sequence having at least 80% identity, preferably at least 90% identity, to the sequence selected from any of SEQ ID NOs: 136, 137, 138, 139, 140, 141, 142, 143, 144, 145, 146, 147, and 148, preferably 143, and wherein the antibody comprises
- 10 (i) a first domain specifically binding IL-17A comprising HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 1, 2, and 3, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 12, 13, and 14, respectively, and
- (ii) a second domain specifically binding TNF α comprising HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 63, 64, and 65, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 76, 77, and 78, respectively, and
- 15 (iii) a third domain specifically binding HSA comprising HCDR1, HCDR2, and HCDR3 sequences of SEQ ID NOs: 90, 91, and 92, respectively, and LCDR1, LCDR2, and LCDR3 sequences of SEQ ID NOs: 100, 101, and 102, respectively, preferably wherein the antibody comprises an amino acid sequence selected from any of SEQ ID NOs: 136, 137, 138, 139, 140, 141, 142, 143, 144, 145, 146, 147, and 148,
- 20 preferably 143.
13. A pharmaceutical composition comprising the multispecific antibody of any one of the preceding claims and a pharmaceutically acceptable carrier.
- 25 14. The multispecific antibody of any one of claims 1 to 12, or the pharmaceutical composition of claim 13 for use as a medicament.
15. A kit comprising the multispecific antibody of any one of claims 1 to 13, or the pharmaceutical composition of claim 14.

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