The question, whether "fully human" mAbs such as derived from phage display libraries or transgenic mice are less immunogenic than humanized mAbs cannot be answered yet, because full immunogenicity data are available for just two human mAbs. An anti-tumor necrosis factor mAb, developed from phage-displayed human libraries induced antibody responses in 12% of patients - at the higher end of the incidence of anti-antibody responses of the humanized antibodies (Hwang, W. Y., et al. (2005). Methods, 36, 3-10).

Evaluation of the immunogenicity of the first registered human mAb generated by the transgenic approach demonstrated that mAb treatment resulted in the generation of antibodies in approximately 5.5% of treated cancer patients (Jakobovits, A., et al. (2007). Nat. Biotechnol. 25, 1134., Lofgren, J. A., et al. (2007). J. Immunol. 178, 7467).

A need therefore remains for a method and means for producing antibodies that are specific for their targets, but are less immunogenic. According to the ~~invention~~disclosure the reduction of immunogenicity is at least partially achieved by providing a transgenic non-human mammal comprising, at least in its B cell lineage, a nucleic acid encoding at least an immunoglobulin light chain or heavy chain, wherein the heavy- or light chain encoding sequence is provided with a means that renders it resistant to DNA rearrangements and/or somatic hypermutations, preferably such a non-human animal is a rodent, more specifically a mouse. The nucleic acid preferably encodes a human, human-like or humanized immunoglobulin chain.

In the remainder of the specification mice are typically used as examples of the non-human mammals. The transgenic non-human mammalian hosts are capable of mounting an immune response to an antigen, where the response produces antibodies having primate, particularly human, variable regions. Various transgenic hosts may be

employed, particularly murine, lagomorpha, ovine, avine, porcine, equine, canine, feline, or the like. Mice have been used for the production of B-lymphocytes for immortalization for the production of antibodies. Since mice are easy to handle, can be bred in large numbers, and are known to have an extensive immune repertoire, mice will usually be the animal of choice. Therefore, in the following discussion, the discussion will refer to mice, but it should be understood that other animals, particularly non-primate mammals, may be readily substituted for the mice, following the same procedures.

The reason for preventing rearrangements and hypermutation is that in this manner a non-immunogenic polypeptide can be chosen beforehand knowing that this polypeptide chain will remain non-immunogenic. At least one of the chains of the resulting immunoglobulin is thus less immunogenic. The resulting antibody needs to have (usually) both a light- and a heavy chain. The non-immunogenic chain must therefore be capable of pairing with the other chain. The other chain may be an endogenous chain, an exogenous chain or a hybrid of both. For human therapy, the non-immunogenic chain should be as close to human as possible.

A means for rendering a gene encoding an immunoglobulin chain (or chains) resistant to DNA rearrangement and/or mutation is of course removal of all genetic elements responsible for said rearrangement and/or mutation. The drawback thereof is that the variability of the two chains is eliminated, whereas the ~~invention~~disclosure preferably retains the variability in one chain (preferably the heavy chain) and inhibits and/or prevents the rearrangement-mutation of the other chain (preferably the light chain).

The elements for rearrangement and/or hypermutation characterized so far are located within the loci for immunoglobulins. Therefore the means for rendering the immunoglobulin encoding sequence resistant to DNA

rearrangement and/or mutation is inserting the gene in a locus outside the immunoglobulin loci.

In a first aspect, there is provided a transgenic murine mammal comprising, at least in its B cell lineage, a nucleic acid molecule encoding a rearranged human immunoglobulin light chain variable region, wherein said light chain variable region is capable of pairing with at least two different heavy chain variable regions encoded by said transgenic, murine mammal, such that variety in specificity of antibodies is retained through rearrangements and hypermutations in the heavy chains, and wherein said light chain further comprises a murine light chain constant region.

Thus according to the ~~invention~~disclosure a transgenic non-human mammal is provided wherein the light/heavy chain encoding sequence is integrated in the genome of the non-human mammal in a locus outside the immunoglobulin loci. Preferably the insertion is in a locus that is resistant to gene silencing. According to the ~~invention~~disclosure, the integration is in the Rosa-locus or a comparable locus.

It is preferred to provide an expression cassette which can be inserted in a Rosa locus or comparable locus with a means that allows expression of the immunoglobulin chain(s) essentially limited to cells of B cell lineage, preferably with a means that allows expression of the light chain encoding nucleic acid during a certain stage of the development of B cells. The term "essentially limited expression" indicates that expression is predominantly in cells of the B-cell lineage, but that lower levels of expression in other cells, as compared to the level of expression in B-cells, is possible. In a preferred embodiment, the term "essentially limited expression" indicates that the expression is exclusively present in cells of the B-cell lineage. Such means typically and preferably include B cell (developmental stage) specific promoters such as CD19, CD20, μHC (all V-

genes), VpreB1, VpreB2, VpreB3, λ5, Igα, Igβ, κLC (all genes), λLC (all genes), BSAP (Pax5). Although it is very well possible to direct the expression of the DNA rearrangement and/or mutation resistant chain by such promoters, they are relatively weak. A strong promoter will typically be required to ensure adequate surface expression of the B cell receptor (made up of the membrane attached Ig H and L chain) and to compete with the expression and pairing of endogenous chains (if present) through allelic exclusion. Such a promoter, however is usually not tissue specific. To confer tissue specificity, an indirect system employing Cre/lox or the like is preferred. The desired chain is put under control of a strong promoter

inhibited by an element that can be removed by the action of a Cre-protein, leading to activation of the desired immunoglobulin encoding gene. This system is described in detail in Wunderlich F. T. (2004), "Generation of inducible Cre systems for conditional gene inactivation in mice", Inauguraldissertation zur Erlangung des Doktorgrades der Mathematisch-Naturwissenschaftlichen Fakultät der Universität zu Köln; http://deposit.ddb.de/cgi-bin/dokserv?idn=97557230x&dok_var=d1&dok_ext=pdf&filename=97557230x.pdf.

Preferably the immunoglobulin chain produced in a manner resistant to rearrangements and hypermutation is a light chain capable of pairing with different heavy chains encoded by the non-human mammal. The light chain will then be the same (and less immunogenic) in all antibodies, but variety in specificity is retained through rearrangements and hypermutations in the heavy chains. It may in that case be preferable to silence at least one of the endogenous loci encoding a light chain, although allelic exclusion may render this unnecessary.

According to this embodiment, preferably the endogenous kappa (κ) light chain locus is functionally silenced.

If the endogenous κ light chain locus is silenced, but also for other reasons, it is preferred that the resistant light chain is a κ light chain, preferably a light chain that has a germline-like sequence. According to the ~~invention~~disclosure such a light chain would lead to an antibody with reduced immunogenicity. The preferred germline sequence is based on the human IGKV1-39 (O12) as this light chain is very frequently observed in the human repertoire (de Wildt et al. 1999. J. Mol. Biol. 285(3):895) and has superior thermodynamic stability, yield and solubility (Ewert et al. 2003. J. Mol. Biol. 325(3):531).

The following gives more specific embodiments of the expression cassette with which the non-human animal can be provided according to the ~~invention~~disclosure. Although this is typically advantageous for immunoglobulins, other genes of interest are also contemplated.

Thus the ~~invention~~disclosure provides in a specific embodiment a transgenic non-human mammal wherein the light chain encoding nucleic acid comprises in 5'-3' direction: a B cell specific promoter, a leader, a rearranged human V gene, optionally a MoEκi enhancer, a constant region (κ) and optionally a (truncated) MoEκ3' enhancer. Neuberger identified and examined a novel B-cell specific enhancer located downstream of the kappa constant region (EP004690251). This enhancer has been shown to play a crucial role in the expression of kappa genes as removal of the 808bp enhancer strongly reduced expression. Deletion of the 3' kappa enhancer also strongly reduced the level of somatic hypermutations (SHM). In transgenic and cell expression studies it has been revealed that reduced, mutated or deleted 3' kappa enhancers not only lowered expression levels but also decreased the level of somatic hypermutations. Currently, it can not be determined whether the 3' kappa enhancer is involved in SHM processes, expression regulation or both (review Odegard, V. H., et al. (2006). Nat. Rev. Immunol. 6, 573; Inlay, M., et al. (2002). Nat. Immunol. 3, 463.).

Detailed expression studies using engineered variants of the 3' kappa enhancer indicated that a 50 nucleotide region is sufficient to drive expression. However for proper expression a reduced sequence of 145 nucleotides is preferred (EP04690251; Meyer, K. B., et al. (1990) Nucleic Acids Res. 18(19):5609-15)

Thus the ~~invention~~disclosure in one ~~aspect~~embodiment provides a nucleic acid for insertion into the genome of a non human animal that is an expression cassette for the expression of a desired proteinaceous molecule in cells developing into

mature B cells during a certain stage of development, said cassette comprising means for preventing silencing of expression of the desired proteinaceous molecule after introduction into a host cell, and means for timing expression of the desired proteinaceous molecule with the desired developmental stage of the host cell.

An expression cassette is defined as a nucleic acid that has been provided with means for introduction into the genome of a host cell, such as sequences which allow for homologous recombination with a certain site in the genome. Usually the nucleic acid will be DNA, typically double stranded. Typically the expression cassette will be provided to the cell in a vector from which it is transferred to the genome of the cell. The expression cassette further comprises all elements necessary for expression of the gene in a host cell, although in certain embodiments some of such elements may be present on a second nucleic acid to be introduced, whereby these elements act in trans. Elements necessary for expression in a host cell include promoters, enhancers and other regulatory elements. Only those elements are necessary that are not provided by the host cell.

According to the ~~invention~~disclosure it is important that the expression of the gene of interest is not silenced in the genome of the host cell, especially not in the development stage where expression is required. This can be done by various means, such as insertion into the endogenous locus or by providing the cassette with nucleic acid elements that prevent silencing (Kwaks et al. (2006) Trends Biotechnol. 24(3), p.137-142; which is incorporated herein by reference). It is preferred that the expression cassette is inserted in a locus that is not silenced in the host cells (EP 01439234; which is incorporated herein by reference).

Said means for prevention of silencing comprise STabilizing Anti-Repression-sequences (STAR®-sequences) and Matrix Attachment Regions (MARs). A STAR sequence is a nucleic acid sequence that comprises a

capacity to influence transcription of genes in cis. Typically, although not necessarily, a STAR sequence does not code by itself for a functional protein element. In one embodiment one STAR element is used. Preferably, however, more than one STAR element is used. In a particularly preferred embodiment an expression cassette as defined above~~according to the invention~~ is provided with two STAR sequences; one STAR sequence at the 5' side of the coding sequence of the immunoglobulin gene and one STAR sequence at the 3' side of the coding sequence of the immunoglobulin gene. MARs are DNA sequences that are involved in anchoring DNA/chromatin to the nuclear matrix and they have been described in both mammalian and plant species. MARs possess a number of features that facilitate the opening and maintenance of euchromatin. MARs can increase transgene expression and limit position-effects.

According to the ~~invention~~disclosure it is important that expression from the cassette only occurs during a certain period in the development of a cell, in particular a developing B cell, more in particular a B cell in a transgenic non-human animal, in particular a mouse. In this particular case the developmental period is chosen such that the expression of the gene from the cassette (typically a light- or heavy chain-like polypeptide) does not significantly interfere with the normal differentiation and/or maturation of the cell and when applicable, allows for pairing of the polypeptide chain produced with its counterpart.

According to the ~~invention~~disclosure this may, in one embodiment, be achieved by providing a nucleic acid as defined above~~according to the invention~~, wherein said means for timing expression is a promoter of which the activity is essentially limited to the certain stage of development. In a developing B cell, which, e.g. after immunization is maturing and/or differentiating, the expression of the gene of interest, when it is one of the polypeptide chains of an immunoglobulin, must not interfere (significantly) with said maturation and/or differentiation and it needs to be timed such that the resulting

polypeptide can pair with its counterparts. Therefore the ~~invention~~disclosure provides a nucleic acid as defined above~~according to the invention~~ wherein said certain stage starts at a stage immediately preceding or coinciding with the onset of the expression of light chain molecules by said cells at a certain stage of development into a mature B cell.

This may be achieved by selecting a promoter which is active only during said suitable period. Such a promoter may be a CD19 promoter, the Ig-α promoter, the Ig-β promoter, the μhc (all genes) promoter, the Vk promoter or analogues or homologues thereof.

In a specific embodiment of the present ~~invention~~disclosure the promoter as disclosed above does not drive the expression of the gene of interest directly. Instead it drives the expression of a gene of which the product activates in trans the expression of the gene of interest. Such an activating gene may be a gene encoding a so-called Cre recombinase or Cre-like protein. The expression cassette for the gene of interest may e.g. be provided with a sequence that inhibits expression of the gene of interest. Said sequence can be removed by the action of the Cre recombinase, which is under control of the desired promoter (active during the proper stage of development). In this embodiment a set of expression cassettes is required.

Therefore the ~~invention~~disclosure provides a set of nucleic acids that are expression cassettes, wherein one nucleic acid comprises an expression cassette encoding a Cre-like protein under control of a promoter active during the desired stage of development of the host cell and the second nucleic acid comprises a sequence encoding a desired proteinaceous molecule under control of a constitutive promoter which can be activated by the action of a Cre-like protein. Said activation is preferably achieved by removal of a stop sequence flanked by loxP sites. The Cre/lox system is described in detail in Rajewsky et al. (1996) J. Clin. Invest. 98, p.600-603, which is incorporated herein by reference. Such systems are reviewed in

Wunderlich F. T. (2004), "Generation of inducible Cre systems for conditional gene inactivation in mice", Inauguraldissertation zur Erlangung des Doktorgrades der Mathematisch-Naturwissenschaftlichen Fakultät der Universität zu Köln; http://deposit.ddb.de/cgi-bin/dokserv?idn=97557230x&dok_var=d1&dok_ext=pdf&filename=975572300x.pd, which is incorporated herein by reference.

The ~~invention~~disclosure further provides a transgenic non-human animal which has been provided with an expression cassette as defined above~~according to invention~~, wherein the desired proteinaceous molecule is a polypeptide chain of an immunoglobulin. A preferred polypeptide chain is a light chain. A more preferred polypeptide is a germline or germline-like light chain. A most preferred polypeptide is O12, preferably the rearranged germline kappa light chain IGKV1-39*01/ IGKJ1*01 (nomenclature according to the IMGT database, http://www.imgt.org).

It is further preferred that the polypeptide chain is rendered essentially incapable of rearrangement and/or of excluded of any sequence modification such as normally operating on Ig during the process of B cell affinity maturation. Therefore, the ~~invention~~disclosure provides a transgenic non-human animal which has been provided with an expression cassette as defined above~~according to the invention~~, wherein said re-arrrangement and/or sequence modifications are prevented by the absence of elements at least partially responsible for somatic hypermutation such as, for example, the MoEκi enhancer.

A preferred expression cassette according to the ~~invention~~disclosure comprises means for prevention of silencing. In one embodiment, said means for prevention of silencing are means for insertion into a locus in the genome of the host cell that is resistant to silencing. Said means for insertion are preferably means for homologous recombination into said site resistant to

silencing. A preferred locus when the non-human animal is a mouse is the rosa-locus.

A further preferred expression cassette according to the ~~invention~~disclosure comprises in 5'-3' direction: a Vκ promoter, a mouse leader, a human V gene, optionally a MoEκi enhancer, a rat constant region (Cκ) and optionally a (truncated) MoEκ3' enhancer.

Yet a further preferred expression cassette according to the ~~invention~~disclosure comprises in 5'-3' direction: a Vκ promoter, a human leader, a human V gene, optionally a MoEκi enhancer, a rat constant region (Cκ) and optionally a (truncated) MoEκ3' enhancer.

Of course the ultimate goal of the invention is to produce antibodies to be used in human therapeutics. Thus in a second aspect, there is~~the invention~~ provide~~s~~d a method for producing a desired antibody comprising exposing a non-human mammal as defined above~~according to the invention~~ to an antigen such that an antibody response is induced and isolating the antibodies specific for the antigen.

In a third aspect, there is~~an alternative embodiment, the invention~~ provide~~s~~d a method for producing a desired antibody comprising exposing a non-human mammal as defined above~~according to the invention~~ to an antigen such that an antibody response is induced and isolating cells producing such antibodies, culturing and optionally immortalizing said cells and harvesting said antibodies.

In a fourth aspect, there is~~further embodiment, the invention~~ provide~~s~~d a method for producing a desired antibody comprising exposing a non-human mammal as defined above~~according to the invention~~ to an antigen such that an antibody response is induced and isolating a nucleic acid encoding at least part of such an antibody, inserting said nucleic acid or a copy or a derivative thereof in an expression cassette and expressing said antibody in a host cell.

The methods for producing antibodies from transgenic mice are known to a person skilled in the art. Particularly preferred are methods for production of mixtures of antibodies from one cell, whereby the nucleic acids encoding these antibodies have been derived from mice according to the ~~invention~~disclosure.

These so-called oligoclonics are disclosed in WO04106375 and WO05068622, which are incorporated herein by reference.

The present ~~invention~~disclosure provides transgenic non-human mammals, preferably mice, capable of generating specific and high affinity hybrid mouse-human antibodies with preferably human immunoglobulin light chain variable (VL) regions in or near germline configuration and preferably murine immunoglobulin heavy chain variable (VH) regions that may have accumulated somatic mutations during the process of antigen-driven affinity maturation. It is envisaged that the murine VH regions of the hybrid antibodies may be subjected to humanization procedures to yield mAbs that have reduced immunogenicity when applied in humans based on germline or near-germline VL regions and murine VH regions that have been humanized.

In particular, it has been shown in the present ~~invention~~disclosure that transgenic mice that harbor a DNA expression construct that encodes a rearranged human VL region under the control of cis-acting genetic elements that provide timely and regulated expression of the transgene on a significant proportion of B cells during B cell development, yet lack elements that direct the somatic hypermutation machinery to the transgene, are capable of generating specific and high affinity mouse-human hybrid antibodies with essentially unmutated L chains. It is shown that the rearranged human transgene is capable of pairing with a diversity of endogenous murine immunoglobulin H chains to form mouse-human hybrid immunoglobulins expressed on the surface of B cells and to

In one embodiment, it is shown that upon immunization with antigen, the B cells are capable of mounting an immune response, leading to the generation of B cells that secrete hybrid antibodies with high specificity and affinity. The V regions encoding these antibodies are characterized by the human transgenic light chain that harbors no or very few mutations and a murine heavy chain that harbors a variable number of mutations introduced by the somatic hypermutation machinery.

In a related embodiment, strategies to obtain high affinity hybrid monoclonal antibodies from the transgenic mice by hybridoma and display technologies are contemplated as well as procedures to humanize the murine VH regions to obtain less immunogenic antibodies for application in humans.

In one embodiment, the ~~invention~~disclosure provides an immunoglobulin L chain transgene construct comprising DNA sequences that encode a human immunoglobulin VL region in combination with a light chain constant region (CL) of an animal immunoglobulin protein, which sequences are operably linked to transcription regulatory sequences that, when integrated in a non-human transgenic animal, produce an Ig VL-CL polypeptide with a human VL region that is not or marginally subject to somatic hypermutation. The Ig VL is capable of pairing with rearranged VH-CH polypeptides that are generated during B cell development in the non-human transgenic animal, with said VH-CH polypeptides retaining the capacity to undergo somatic hypermutation upon stimulation. The CL region may be of any animal species and is generally capable of pairing with the CH regions of the non-human transgenic animal.

The ~~invention~~disclosure also includes the use of a transgene construct as above in producing a transgenic non-human animal capable of the production of hybrid antibodies consisting of VL-CL polypeptides and VH-CH polypeptides in which the VL region is of human origin and the CL, VH and

CH may be of any animal species, including human. Upon immunization, these transgenic animals are capable of generating high affinity antibodies encoded by somatically hypermutated VH genes and essentially non-mutated VL genes encoded by the transgene.

In another ~~aspect~~embodiment, the ~~invention~~disclosure provides a process for the production of a transgenic non-human animal capable of the production of hybrid antibodies in response to antigenic challenge, comprising functionally disrupting the endogenous immunoglobulin light chain locus and inserting into the animal genome a transgene construct of the ~~invention~~disclosure.

The ~~invention~~disclosure includes the use of animals obtainable by this process in the production of B cells that produce immunoglobulin having human VL light chain. In another ~~aspect~~embodiment of the ~~invention~~disclosure there is provided a process for the production of B cells that produce immunoglobulin having human VL and binding to a selected antigen, comprising challenging an animal obtainable by a process as above with said antigen and screening for B cells from said animal that bind said antigen. The ~~invention~~disclosure further includes B cells obtainable by this process and hybridomas obtainable by immortalizing such B cells, e.g. hybridomas obtained by fusing B cells as above with myeloma cells. The ~~invention~~disclosure also includes a process for producing monoclonal antibody comprising cultivating such a hybridoma. In yet a further ~~aspect~~embodiment, the ~~invention~~disclosure provides the use of the above B cells in producing a hybridoma or corresponding monoclonal antibody.

Yet a further ~~aspect~~embodiment of the ~~invention~~disclosure is a process for the production of immunoglobulin having human VL chain and binding to a selected antigen, comprising challenging an animal obtainable as above with said antigen and obtaining immunoglobulin there from.

In one strategy, as an individual step, a rearranged VL region encoded by human germline V and J gene segments and a light chain

transgene to increase expression of the transgene in animals, preferably in B-cells.

The ~~invention~~disclosure further provides progeny of a transgenic non-human animal as defined above~~according to the invention~~, the progeny comprising, at least in its B-cell lineage, a heavy- or light chain encoding sequence together with a means that renders the sequence resistant to DNA rearrangements and/or somatic hypermutations.

The ~~invention~~disclosure further provides progeny of a transgenic non-human animal as defined above~~according to the invention~~, the progeny comprising an expression cassette for the expression of a desired proteinaceous molecule in cells during a certain stage of development in cells developing into mature B cells.

The ~~invention~~disclosure in addition provides a cell that is isolated from a transgenic non-human animal as defined above~~according to the invention~~, the cell comprising a heavy- or light chain encoding sequence together with a means that renders the sequence resistant to DNA rearrangements and/or somatic hypermutations. The ~~invention~~disclosure in addition provides a cell that is isolated from a transgenic non-human animal as defined above~~according to the invention~~, the cell comprising an expression cassette for the expression of a desired proteinaceous molecule in cells during a certain stage of development in cells developing into mature B cells. A cell according to the ~~invention~~disclosure, preferably an antibody-producing B-cell or a cell that is capable of differentiating or maturating into an antibody-producing B-cell, can be used for *in vitro* production of antibodies, as is known to the skilled person, for example from Gascan et al. 1991. J. Exp. Med. 173: 747-750. Methods for immortalization of a cell according to the ~~invention~~disclosure are known in the art and include the generation of hybridomas, for example by fusion with a myeloma cell, transformation with Epstein Barr Virus; expression of the signal transducer of activation and transcription (STAT), activation via

applied in humans and result in more stable antibodies that are less prone to aggregation and thus safer for therapeutic use in humans.

MAbs derived from the afore-mentioned non-human transgenic animals or cells all share the same identical human VL regions. It has been described that mAbs that share the same identical VL region may be co-expressed in a single clonal cell for the production of mixtures of recombinant antibodies with functional binding sites (see WO04106375 and WO05068622). Thus, the ~~invention~~disclosure provides a platform for the generation of specific and high affinity mAbs that constitute the basis for mixtures of mAbs produced by clonal cells.

It is preferred that mAbs derived from the afore-mentioned non-human transgenic animals or cells are directed against cellular targets. Preferred targets are human surface-expressed or soluble proteins or carbohydrate molecules. Further preferred targets are surface-expressed proteins or carbohydrate molecules that are expressed on the surface of bacteria, viruses, and other pathogens, especially of humans.

More specifically, preferred targets include cytokines and chemokines, including but not limited to InterLeukin 1beta (IL1beta), IL2, IL4, IL5, IL7, IL8, IL12, IL13, IL15, IL18, IL21, IL23 and chemokines such as, for example, CXC chemokines, CC chemokines, C chemokines (or γ chemokines) such as XCL1 (lymphotactin-α) and XCL2 (lymphotactin-ß), and CX3C chemokines. Further included as preferred targets are receptor molecules of the cytokines and chemokines, including type I cytokine receptors such as, for example, the IL-2 receptor, type II cytokine receptors such as, for example interferon receptors, immunoglobulin (Ig) superfamily receptors, tumor necrosis factor receptor family including receptors for CD40, CD27 and CD30, serine/threonine-protein kinase receptors such as TGF beta receptors, G-protein coupled receptors such as CXCR1-CXCR7, and tyrosine kinase receptors such as fibroblast growth factor receptor

virus, and Varicella zoster virus, parvoviruses such as, for example, B19; Legionella pneumophila; Listeria monocytogenes; Campylobacter jejuni; Staphylococcus aureus; E. coli O157:H7; Borrelia burgdorferi; Helicobacter pylori; Ehrlichia chaffeensis; Clostridium difficile; Vibrio cholera; Salmonella enterica Serotype Typhimurium; Bartonella henselae; Streptococcus pyogenes (Group A Strep); Streptococcus agalactiae (Group B Strep); Multiple drug resistant S. aureus (e.g. MRSA); Chlamydia pneumoniae; Clostridium botulinum; Vibrio vulnificus; Parachlamydia pneumonia; Corynebacterium amycolatum; Klebsiella pneumonia; Linezolid-resistant enterococci (E. faecalis and E. faecium); and Multiple drug resistant Acinetobacter baumannii.

Most preferred targets are IL-6 and its receptor, IL-6Ralpha, glycoprotein-denominated gp130, RSV, especially the surface proteins F, G and SH and non-structural proteins such as N and M, and receptor tyrosine kinases, in particular erbB1 (EGF-R; HER1), erbB2, (HER2), erbB3 (HER3), erbB4 (HER4), IGF-R1 and IGF-RII, c-Met (HGF-R).

Therefore, the ~~invention~~disclosure provides a platform for the generation of specific and high affinity mAbs against the above mentioned targets that constitute the basis for mixtures of mAbs produced by clonal cells. In a preferred embodiment, said specific and high affinity mAbs comprise mAbs that are directed against different epitopes on at least one of the targets. In a further preferred embodiment, said specific and high affinity mAbs comprise mAbs that are directed against different targets, such as, for example, one or more members of the EGF-receptor family, including erbB1 (EGF-R; HER1), erbB2, (HER2), erbB3 (HER3) and erbB4 (HER4).

Unless otherwise defined, scientific and technical terms used in connection with the present ~~invention~~disclosure shall have the meanings that are commonly understood by those of ordinary skill in the art. Further, unless otherwise required by context, singular terms shall include pluralities and