

4-1BBL is the activating ligand of the 4-1BB receptor (CD137), a member of the TNF receptor superfamily and a potent activation-induced T cell costimulatory molecule. 4-1BBL naturally forms a homo-trimer but signaling via 4-1BB requires significant oligomerization of 4-1BBL. 4-1BBL is present on a variety of antigen presenting cells (APCs), including dendritic cells (DCs), B cells, and macrophages. The 4-1BB receptor is not detected (<3%) on resting T cells or T cell lines, however, 4-1BB is stably upregulated when T cells are activated. 4-1BB activation upregulates survival genes, enhances cell division, induces cytokine production and prevents activation induced cell death in T-cells.

Additional background art includes:

Weiskopf K et al. Science. (2013); 341(6141):88-91;
 International Patent Application Publication No. WO 2017027422;
 International Patent Application Publication No. WO 2001086003;
 International Patent Application Publication No. WO 2001075067;
 International Patent Application Publication No. WO 2017194641;
 International Patent Application Publication No. WO 2014180288;
 International Patent Application Publication No. WO2017059168;
 International Patent Application Publication No. WO2001/049318;
 International Patent Application Publication No. WO2016/139668;
 International Patent Application Publication No. WO2014/106839;
 International Patent Application Publication No. WO2012/042480;
 US Patent Application Publication No. 20150183881;
 US Patent Application Publication No. US20070110746;
 US Patent Application Publication No. US20070036783; and
 US Patent No. US9562087.

SUMMARY

In one aspect, the present disclosure refers to a SIRP α -4-1BBL fusion protein comprising a SIRP α amino acid sequence and a 4-1BBL amino acid sequence, wherein: (a) said SIRP α amino acid sequence is as set forth in SEQ ID NO: 2 and said 4-1BBL amino acid sequence is as set forth in SEQ ID NO: 23; (b) said SIRP α amino acid sequence is as set forth in SEQ ID NO: 24 and said 4-1BBL amino acid sequence is as set forth in SEQ ID NO: 22; or (c) said SIRP α amino acid sequence is as set forth in SEQ ID NO: 2 and said 4-1BBL amino acid sequence is as set forth in SEQ ID NO: 22; and wherein said fusion protein is capable of at least one of:

- (i) binding CD47 and 4-1BB;
- (ii) activating said 4-1BB signaling pathway in a T cell expressing said 4-1BB;

- (iii) co-stimulating T cells expressing said 4-1BB; and/or
- (iv) enhancing phagocytosis of cancer cells expressing said CD47 by phagocytes selected from the group consisting of macrophages and granulocytes compared to same in the absence of said SIRP α -4-1BBL fusion protein.

According to an example, the present disclosure refers to a SIRP α -4-1BBL fusion protein comprising a SIRP α amino acid sequence and a 4-1BBL amino acid sequence, wherein the SIRP α amino acid sequence is 100 - 119 amino acids in length having at least 95 % identity to an amino acid sequence selected from the group consisting of SEQ ID NOs: 24 and 26, and/or wherein the 4-1BBL amino acid sequence:

- (a) is 185 - 202 amino acids in length having at least 95 % identity to an amino acid sequence selected from the group consisting of SEQ ID NOs: 22, 23, 27 and 28, is 170 – 197 amino acids in length having at least 80 % identity to an amino acid sequence selected from the

According to an example of the present disclosure there is provided a method of activating immune cells, the method comprising in-vitro activating immune cells in the presence of the SIRP α -4-1BBL fusion protein or the isolated polypeptide, the polynucleotide or the nucleic acid construct or the host cell. According to an aspect, the present disclosure refers to an *in-vitro* method of activating immune cells, the method comprising in-vitro activating immune cells in the presence of the SIRP α -4-1BBL fusion protein or the isolated polypeptide, the polynucleotide or the host cell as disclosed herein.

According to some examples of the disclosure, the activating is in the presence of cells expressing CD47 or exogenous CD47.

According to some examples of the disclosure, the cells expressing the CD47 comprise pathologic cells.

According to some examples of the disclosure, the pathologic cells comprise cancer cells.

According to some examples of the disclosure, the activating is in the presence of an anti-cancer agent.

According to some examples of the disclosure, the therapeutic agent for treating the disease or the anti-cancer agent comprises an antibody.

According to some examples of the disclosure, the antibody is selected from the group consisting rituximab, cetuximab, trastuzumab, edrecolomab, almetuzumab, gemtuzumab, ibritumomab, panitumumab, Belimumab, Bevacizumab, Bivatuzumab mertansine, Blinatumomab, Blontuvetmab, Brentuximab vedotin, Catumaxomab, Cixutumumab, Daclizumab, Adalimumab, Bezlotoxumab, Certolizumab pegol, Citatuzumab bogatox, Daratumumab, Dinutuximab, Elotuzumab, Ertumaxomab, Etaracizumab, Gemtuzumab ozogamicin, Girentuximab, Necitumumab, Obinutuzumab, Ofatumumab, Pertuzumab, Ramucirumab, Siltuximab, Tositumomab, Trastuzumab, Nivolumab, Pembrolizumab, Durvalumab, Atezolizumab, Avelumab and ipilimumab.

According to some examples of the disclosure, the antibody is selected from the group consisting of rituximab, cetuximab and almetuzumab.

According to some examples of the disclosure, the therapeutic agent for treating the disease or the anti-cancer agent comprises an IMiD (e.g. Thalidomide, Lenalidomide, Pomalidomide).

According to some examples of the disclosure, the method comprising adoptively transferring the immune cells following the activating to a subject in need thereof.

According to some examples of the disclosure, the immune cells comprise T cells.

According to some examples of the disclosure, the immune cells comprise phagocytes.

WHAT IS CLAIMED IS:

1. A SIRP α -4-1BBL fusion protein comprising a SIRP α amino acid sequence and a 4-1BBL amino acid sequence, wherein:
 - (a) said SIRP α amino acid sequence is as set forth in SEQ ID NO: 2 and said 4-1BBL amino acid sequence is as set forth in SEQ ID NO: 23;
 - (b) said SIRP α amino acid sequence is as set forth in SEQ ID NO: 24 and said 4-1BBL amino acid sequence is as set forth in SEQ ID NO: 22; or
 - (c) said SIRP α amino acid sequence is as set forth in SEQ ID NO: 2 and said 4-1BBL amino acid sequence is as set forth in SEQ ID NO: 22;and
wherein said fusion protein is capable of at least one of:
 - (i) binding CD47 and 4-1BB;
 - (ii) activating said 4-1BB signaling pathway in a T cell expressing said 4-1BB;
 - (iii) co-stimulating T cells expressing said 4-1BB; and/or
 - (iv) enhancing phagocytosis of cancer cells expressing said CD47 by phagocytes selected from the group consisting of macrophages and granulocytes compared to same in the absence of said SIRP α -4-1BBL fusion protein.
2. The SIRP α -4-1BBL fusion protein of claim 1, wherein said SIRP α -4-1BBL fusion protein amino acid sequence comprises an amino acid sequence selected from the group consisting of SEQ ID NO: 11, 13 and 16.
3. The SIRP α -4-1BBL fusion protein of claim 1, wherein said SIRP α -4-1BBL fusion protein amino acid sequence consists of an amino acid sequence selected from the group consisting of SEQ ID NO: 11, 13 and 16.
4. A polynucleotide encoding the SIRP α -4-1BBL fusion protein or the polypeptide of any one of claims 1-3.
5. A host cell comprising the SIRP α -4-1BBL fusion protein or the polypeptide of any one of claims 1-3 or the polynucleotide of claim 4.

6. A method of producing a SIRP α -4-1BBL fusion protein or a polypeptide, the method comprising expressing in a host cell the polynucleotide of claim 4.

7. Use of the SIRP α -4-1BBL fusion protein of any one of claims 1-3, the polynucleotide of claim 4 or the host cell of claim 5, in the manufacture of a medicament for treating cancer.

8. An *in-vitro* method of activating immune cells, the method comprising in-vitro activating immune cells in the presence of the SIRP α -4-1BBL fusion protein or the isolated polypeptide of any one of claims 1-3, the polynucleotide of claim 4 or the host cell of claim 5.