CLAIMS

1. A method of identifying a cell which produces a scFv which is a specific binding partner for a target polypeptide, the method comprising the steps:

(a) expressing a library of scFvs in a population of mammalian cells,

wherein each cell in the population of mammalian cells expresses at least one member of the scFv library from one or more retroviral (e.g. lentiviral) vectors which have been integrated into the genome of that cell,

wherein each scFv comprises a first detectable tag,

wherein each scFv is secreted from the cell in the population of cells in which it is produced,

wherein each cell in the population of mammalian cells comprises an expression construct comprising an inducible promoter operably linked to a nucleotide sequence encoding the target polypeptide, wherein the target polypeptide is an integrated membrane protein, wherein the target polypeptide comprises a second detectable tag, and wherein the expression construct is integrated into the genome of each cell;

(b) optionally removing cells from the population of mammalian cells to which scFvs bind;

(c) inducing expression of the target polypeptide from the expression construct such that the target polypeptide is displayed on the outer surface of each cell in the population of mammalian cells; and

(d) isolating cells within the population of mammalian cells to which secreted scFvs which are specific binding partners for the target polypeptide are bound to the target polypeptide, wherein specific binding partners for the target polypeptide are ones which bind to the target polypeptide though at least one antigen recognition site located in a variable region of the scFv,

wherein cells to which scFvs are bound are isolated using magnetic activated cell sorting (MACS),

detecting the first and second detectable tags using independent antibodies to which are attached first and second members of a donor-acceptor FRET pair,

wherein the cells on which scFvs are detected are cells which produce scFvs which bind to the target polypeptide.

2. The method as claimed in claim 1, wherein the method additionally comprises, prior to Step (a), the step of:

contacting the population of mammalian cells with a library of retroviral (e.g. lentiviral) particles, each particle comprising a retroviral (e.g. lentiviral) vector encoding a member of a scFv library and a detectable tag, under conditions such that at least one retroviral (e.g. lentiviral) vector is integrated into the genome of each cell (or substantially each cell) in the library.

3. The method as claimed in clam 1 or claim 2, wherein the method additionally comprises, prior to Step (a), the step:

integrating, into the genome of each (or substantially each) cell in the population of cells, an expression construct comprising an inducible promoter) operably linked to a nucleotide sequence encoding the target polypeptide and wherein the target polypeptide comprises a second detectable tag.

4. The method as claimed in any one of the preceding claims, wherein the detectable tag is HA or a Myc tag.

5. The method as claimed in any one of the preceding claims, wherein the inducible promoter comprises a plurality of Tet operator sequences to which the Tet repressor protein (TetR) is capable of binding.

6. The method as claimed in any one of the preceding claims, wherein Step (b) and/or Step (c) additionally comprises the step:

contacting all or part of the population of mammalian cells with an excess of capturing cells which express the target polypeptide (and optionally a second detectable tag), but do not express a scFv,

wherein each capturing cell comprises a label (preferably a fluorescence label) which allows the capturing cells to be distinguished from the population of mammalian cells.

7. The method as claimed in any one of the preceding claims, wherein the first and second members of the donor-acceptor FRET pair are FITC and Alexa Fluor 594.

8. The method as claimed in any one of the preceding claims, wherein the method additionally comprises the step:

(e) sequencing part of all of the nucleotide sequences in the isolated cells or identified cells which encode the scFvs which bind to the target polypeptide, preferably using a non-Sanger sequencing method.