Claims

1. A method for identifying a biomarker indicative of a reduced response to a drug in a patient comprising the steps of :
    a) heating either i) a sample from a patient having a reduced response to the drug or ii) a sample from a patient responsive to the drug;
    b) separating soluble from insoluble protein of i) or ii); and
    c) analysing either or both the soluble and insoluble fractions of step b) and either comparing with the corresponding fraction obtained from heating and separating a sample from a patient responsive to the drug when i) is heated and separated in steps a) and b) , or comparing with the corresponding fraction obtained from heating and separating a sample from a patient having a reduced response to the drug when ii) is heated and separated in steps a) and b),

    to identify at least one protein which has a different melting temperature in the sample from the reduced drug response sample, compared to the sample from drug responsive patient.

2. A method for identifying a biomarker indicative of a reduced response to a drug in a patient comprising the steps of
    a) heating a sample from a patient responsive to the drug,
    b) separating soluble from insoluble protein in the product of step a),
    c) analysing either or both the soluble and insoluble protein fractions of step b),
    d) repeating steps a) to c) with a sample from a patient having a reduced response to the drug, and
    e) identifying at least one protein which has a different melting temperature in the sample from the drug responsive patient compared to the sample from the reduced drug responsive patient, wherein said protein is a biomarker indicative of a reduced response to the drug,

    wherein step d) is optionally carried out prior to or simultaneously to steps a) to c).

3. The method of claim 1 or 2 wherein said patient has been treated with said drug.

4. The method of any one of claims 1 to 3 wherein step c) determines the melting

temperature of one or more proteins.

5. The method of any one of claims 1 to 4 wherein the soluble or insoluble fraction of step b) is analysed by mass spectrometry.

6. The method of any one of claims 1 to 5 wherein the target protein for the drug is excluded from identification.

7. A method for generating a proteome profile from a sample, indicative of a reduced response to a drug in a patient comprising the steps of:
   a) heating a sample from a patient responsive to the drug,
   b) separating soluble from insoluble protein,
   c) analysing either or both the soluble and insoluble protein fractions of step b) to determine the melting temperature of a plurality of proteins,
   d) repeating steps a) to c) with a sample from a patient having a reduced response to the drug, and
   e) producing a proteome profile for the plurality of proteins indicating quantitative differences between the melting temperatures obtained for proteins from the sample from a patient responsive to the drug and the melting temperatures obtained for proteins from the sample from a patient with a reduced response to the drug,
   wherein optionally step d) may be performed prior to or simultaneously to steps a) to c).

8. The method of any one of claims 1 to 7 wherein said sample is a tissue sample.

9. The method of claim 8 wherein said tissue sample is blood, serum, plasma, lymph, cerebrospinal fluid, mucus, urine, faces or is a liquid or solid tumour sample.

10. The method of any one of claims 1 to 9 wherein said sample from a patient responsive to the drug and said sample from a patient having a reduced response to the drug are of the same type.

11. The method of any one of claims 1 to 10 wherein said sample from a patient responsive to the drug and said sample from a patient with a reduced response to the drug are taken from the same patient at different times.

12. The method of any one of claims 1 to 11 wherein a step of cell sorting is carried out before said analysis step.

13. The method of any one of claims 1 to 12 further comprising a lysis step.

14. The method of claim 14 wherein said lysis step is carried out after step a).

15. The method of anyone of claims 1 to 14 wherein the samples are exposed to a series of different temperatures.

16. The method of any one of claims 1 to 15 wherein said separation step is carried out by filtration, centrifugation or affinity capture of soluble protein.

17. A method for determining whether a protein is a biomarker indicative of a reduced response to a drug in a patient, comprising the steps of:
    a) heating a sample from a patient responsive to the drug,
    b) analysing the product of step a) for the presence of the soluble form of said protein using at least two affinity reagents which are capable of binding to the soluble protein with a higher affinity than to an unfolded or insoluble form of the protein,
    c) repeating steps a) and b) with a sample from a patient having a reduced response to the drug, and
    d) determining whether there is any difference in the amount of soluble protein present in the drug responsive and reduced drug response samples, wherein if a difference is detected, the protein is a biomarker indicative of a reduced response to a drug;
    wherein optionally, step c) is carried out prior to or simultaneously to steps a) and b).