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(54) Title: BIOMARKERS PREDICTING SENSITIVITY TO PP2A ACTIVATION THERAPY IN CANCER

(57) Abstract: The invention relates to the treatment and classification of cancer, in particular breast cancer. It relates to identifying patients who are likely to respond to cancer therapy with a PP2 A activator.

BIOMARKERS PREDICTING SENSITIVITY TO PP2A ACTIVATION THERAPY IN CANCER

The invention relates to the treatment and classification of cancer, in particular breast cancer. It relates to identifying patients who are likely to respond to
5 cancer therapy with a PP2A activator.

BACKGROUND

The provision of new treatments for cancer is of high importance, including for cancers resistant to known treatments. There is also a need for identification of markers
10 that allow for detection, prognosis and classification of cancer, and which can predict responsiveness of a patient to a given therapy.

The deregulation of the protein phosphatase 2A (PP2A) complex is known to be a common event in cancer (Grech et al; Tumor Biol; DOl 10.1007/s13277-016-5145-4 (2016)).

15
SUMMARY OF INVENTION

The inventors have shown that cancer cells sensitive to treatment with a PP2A activator exhibit overexpression of the markers AURKA and KIF2C. These include cancer cells exemplified by Triple negative breast cancer (TNBC) cells that do not benefit from targeted therapy and have bad prognosis when current front-line
20 anti-cancer therapies are administered. The markers further correlate with low PP2A enzymatic activity.

The markers may thus be used to predict responsiveness of a patient to treatment with a PP2A activator. AURKA and KIF2C may also be targeted with antagonists to thereby treat cancer, based on their correlation with cancer.

25 In addition, the inventors have also shown that the markers AURKA and KIF2C have utility in classifying cancer. The markers may be used to classify cancers with a worse prognosis, in particular basal versus luminal breast cancer subtype. The markers may thus be assayed using suitable reagents in kits for classification of cancer and for predicting therapeutic benefit from activation of the PP2A
30 complex.

The invention therefore provides a method of treating a patient having a cancer comprising an overexpression of AURKA and/or KIF2C, comprising administering to the patient a PP2A activator and thereby treating the cancer. The invention additionally provides a method of treating cancer in a patient, the method comprising (a) measuring the amount of AURKA and/or KIF2C in the cancer and (b) if the cancer comprises an overexpression of AURKA and/or KIF2C, administering to the patient a PP2A activator and thereby treating the cancer.

The invention also provides a PP2A activator for use in treating cancer in a patient, wherein the cancer comprises an overexpression of AURKA and/or KIF2C, and use of a PP2A activator in the manufacture of a medicament for treating cancer in a patient, wherein the cancer comprises an overexpression of AURKA and/or KIF2C.

The invention also provides a method for determining whether or not a patient having or suspected of having or being at risk of developing cancer will respond to treatment with a PP2A activator, which method comprises measuring expression of AURKA and/or KIF2C in the individual, and thereby predicting whether or not the patient will respond to treatment with a PP2A activator.

The invention further provides a method for classifying a cancer in a patient, the method comprising measuring expression of AURKA and/or KIF2C in the patient, and classifying the cancer as of a particular subtype based on the expression. The invention also provides a kit for detecting a cancer comprising a deregulation of PP2A, comprising reagents suitable for detecting expression of AURKA and/or KIF2C. The invention also provides a system for classifying cancer in a patient, or for predicting responsiveness of a cancer patient to treatment with a PP2A activator, the system comprising: (a) a measuring module for determining expression of AURKA and/or KIF2C in the patient, (b) a storage module configured to store control data and output data from the measuring module, (c) a computation module configured to provide a comparison between the value of the output data from the measuring module and the control data; and (d) an output module configured to display whether or not the patient has cancer based on the comparison, wherein an overexpression of AURKA and/or KIF2C in the patient classifies the cancer or predicts that the patient will respond to treatment with a PP2A activator.

BRIEF DESCRIPTION OF FIGURES

Figure 1 shows MTT assays for cell viability of various breast cancer cell lines at increasing concentrations of FTY720 ranging from 0 (untreated) to 5μM. Cells were left to adhere for 24 hours following seeding, then treated for 48 hours in duplicate experiments. The cell viability of the cell lines at each dose is expressed as a percentage of the untreated cells. Error bars are expressed as percentages of the untreated cell viability and represent the standard deviation from triplicate assay values for biological replicates. [A] Sensitive basal and triple negative breast cancer (TNBC) cell lines; [B] Basal and triple negative cell lines that are not sensitive to FTY720; [C] Normal-like cell lines: HB-2 and MCF10A; [D] MCF-7, MDAMB453 Luminal, ER positive cell lines; and [E] Luminal, HER2 positive SKBR-3 and BT-474.

Figure 2. Distribution of gene expression levels derived from quantigene assay intensity data, normalised to housekeeping genes, comparing sensitive versus non sensitive cell lines. RNA expression was measured by Quantigene and normalised to housekeeping gene expression of each cell line respectively. [A] Expression levels of PP2A complex subunits: PPP2C A (white), PPP2R2 A (hatched) and the inhibitory subunit,Cip2a (black). The adjacent table shows the statistical significance of the difference in the median expression between the two categories of cell lines that are sensitive or not sensitive to FTY720 [* shows significant P-values to 95% confidence interval]. [B] expression levels of AURKA (black) and KIF2C (white). The adjacent table shows a statistically significant difference in expression between sensitive and non-sensitive cell lines for AURKA and KIF2C gene expression. Statistical analysis was done using the Mann-Whitney U test and a p-value smaller than 0.05 was considered significant. The expression of Cip2a, AURKA and KIF2C are significantly higher in the FTY720-senstive cell lines (p values of <0.05, <0.02, <0.001 respectively). Sensitive cell lines thus include the TNBC cell lines MDAMB231, BT-20 and Hs578T, while non-sensitive cell lines include HCC1937, MDAMB436, MDAMB468, MDAMB453, BT-474, MCT-7 and SKBR-3.

Figure 3. AURKA (white) and KIF2C (black) expression levels derived from RNASeq dataset downloaded from TCGA data portal (n=72), *https://portal.gdc.cancer.gov/ onl 3 January 2014.* [A] Distribution of AURKA and KIF2C

expression across breast cancer subtypes as defined by the PAM50 annotation, compared to normal tissues. [Normal N = 82; Basal N = 97; HER2-Enriched N = 82; Luminal B N = 129; Luminal A N = 228][B] Comparison of expression in patient tumour tissue and matched normal tissue. The statistical significance of the differential expression between tumours and matched normal tissue is shown in the adjacent table using the Related samples Wilcoxon signed rank test. The tumour versus normal column shows the direction of significant difference where applicable. [n = 72; RNASeqV2 normalised expression as downloaded from TCGA data portal]. [C] Comparison of expression in patient tumour tissue and matched normal tissue across different PAM50 defined patient subtypes (N = 55).

Figure 4. Percent patients with amplifications or an expression level (z-score) greater than 2 (AMP; EXP>2) for AURKA (white bars) and KIF2C (black bars) in tumours of different origin. The analysis of data was done using the data portal (cBioPortal for Cancer Genomics (Cerami et al., 2012), available at http://www.cbioportal.org. The normalised RNASeqV2 data was used from the TCGA dataportal (https://tcga-data.nci.nih.gov/tcga/).

Description of the Sequence Listing

SEQ ID NO: 1 is a DNA sequence for AURKA.

SEQ ID NO:2 is an amino acid sequence for AURKA.

SEQ ID NO: 3 is a DNA sequence for KIF2C.

SEQ ID NO:4 is an amino acid sequence for KIF2C.

DETAILED DESCRIPTION

5 It is to be understood that different applications of the disclosed products and methods may be tailored to the specific needs in the art. It is also to be understood that the terminology used herein is for the purpose of describing particular embodiments of the invention only, and is not intended to be limiting.

In addition as used in this specification and the appended claims, the singular forms

"a", "an", and "the" include plural referents unless the content clearly dictates otherwise. Thus, for example, reference to "an inhibitor" includes two or more such inhibitors, or reference to "an oligonucleotide" includes two or more such oligonucleotide and the like.

All publications, patents and patent applications cited herein, whether supra or infra, are hereby incorporated by reference in their entirety.

Methods of Treatment

The invention provides a method of treating a patient having a cancer comprising an overexpression of AURKA and/or KIF2C, comprising administering to the patient a PP2A activator and thereby treating the cancer.

Cancer

The cancer may be any cancer or tumour. Deregulation (low activity) of the PP2A complex is observed in a wide range of cancers (as described in Grech et al, supra). Thus, identification of markers whose overexpression correlates with low PP2A activity provides for treatment of a patient displaying such overexpression with a PP2A activator.

Particular cancers and tumours that may be treated include stomach, colorectal, breast and pancreatic cancers and tumours, based on the high expression of AURKA and/or KIF2C in such cancers in the TCGA (The Cancer Genome Atlas) dataset according to the inventors' analysis (FIGURE 4). A particularly preferred cancer or tumour for treatment is breast cancer or a breast tumour. An especially preferred form of breast cancer for treatment is basal breast cancer.

Markers

The cancer selected for treatment preferably has an overexpression of AURKA. The overexpression of AURKA may be present without an overexpression of KIF2C. The cancer may have an overexpression of both AURKA and KIF2C. AURKA is an abbreviation for aurora kinase A, and is additionally referred to in the art as AIK; ARJ1; ARUA; BTAK; STK6; STK7; STK15; PPP1R47. A sequence for AURKA is provided at NM_198433.2- all references to accession numbers herein relate to the sequence provided as of 22 March 2017. An AURKA nucleic acid sequence preferably comprises the

sequence shown in SEQ ID NO: 1 or a variant thereof. An AURKA nucleic acid sequence may comprise the sequence of a transcript variant of SEQ ID NO: 1, such as the sequence of any one of NM_003600.3, NM_198434, NM_198435.2, NM_198436.3, NM_001323303.1, NM_001323304.1 or NM_001323305.1 or a variant of any thereof. An
5 AURKA protein preferably comprises the sequence shown in SEQ ID NO: 2 or a variant thereof.

The cancer may have an overexpression of KIF2C. The overexpression of KIF2C may be present without an overexpression of AURKA. KIF2C is an abbreviation for kinesin family member 2C, and is additionally referred to in the art as MCAK; CT139;
10 KNSL6. A sequence for KIF2C is provided at NM_006845.3. AKIF2C nucleic acid sequence preferably comprises the sequence shown in SEQ ID NO: 3 or a variant thereof. . An AURKA nucleic acid sequence may comprise the sequence of a transcript variant of SEQ ID NO: 3, such as the sequence of any one of NM_00 129765 5.1, NM_001297656.1, or NM_001297657.1 or a variant of any thereof. AKIF2C protein preferably comprises the
15 sequence shown in SEQ ID NO: 4 or a variant thereof. A KIF2C protein may comprise the sequence encoded by any of the above transcript variants of SEQ ID NO:3, or a variant thereof.

Over the entire length of the amino acid sequence of SEQ ID NO: 2 or 4, the variant will preferably be at least 90% homologous to that sequence based on amino acid
20 identity, i.e. have at least 90% amino acid identity over the entire sequence. More preferably, the variant may be at least 95%, 97% or 99% homologous based on amino acid identity (or identical) to the amino acid sequence of SEQ ID NO: 2 or 4 over the entire sequence. Over the entire length of the sequence of SEQ ID NO: 1 or 3, a variant will preferably be at least 90% homologous to that sequence based on nucleotide identity over
25 the entire sequence, i.e. have at least 90% nucleotide identity over the entire sequence. More preferably, the variant may be at least 95%, 97% or 99% homologous based on nucleotide identity (or identical) to the nucleotide sequence of SEQ ID NO: 1 or 3 over the entire sequence.

Standard methods in the art may be used to determine homology. For example the
30 UWGCG Package provides the BESTFIT program, which can be used to calculate homology, for example used on its default settings (Devereux et al (1984) Nucleic Acids

Research 12, p387-395). The PILEUP and BLAST algorithms can be used to calculate homology or line up sequences (such as identifying equivalent residues or corresponding sequences (typically on their default settings)), for example as described in Altschul S. F. (1993) J Mol Evol 36:290-300; Altschul, S.F et al (1990) J Mol Biol 215:403-10. Software
5 for performing BLAST analyses is publicly available through the National Center for Biotechnology Information (http://www.ncbi.nlm.nih.gov/).

The cancer selected for treatment may have an overexpression or lack overexpression of other markers. A cancer that originates from breast, may have lack of overexpression ("negative") for one or more of FIER2, ER and PR, such as FIER2 and ER,
10 HER2 and PR, ER and PR, PR and HER2, or HER2, ER and PR. These three markers are key targets for therapeutic intervention in breast cancer and thus cancers negative for these markers have a worst prognosis, lacking targeted therapy. .The present inventors have shown that AURKA and/or KIF2C may be overexpressed in breast cancer negative for the three markers ("triple negative"), and thus provide a new avenue for treatment of a subtype
15 of these cancers, with a PP2A activator or an antagonist of AURKA or KTF2C.

Alternatively, the cancer may have an overexpression of FIER2. The inventors have shown that FIER2 positive cancers may also display an overexpression of AURKA (Table 2). Both triple negative breast cancers (TNBC) and Her2 positive breast cancers are aggressive tumours which may reflect a requirement for AURKA expression during
20 mitosis (Afonso et al 2016. Interestingly, KIF2C is overexpressed almost exclusively in the Basal subtype of breast tumours, highly represented by TNBC patients (Table 2) and thus provides a marker for basal breast tumours

The cancer may have an overexpression of one or more endogenous PP2A inhibitors. The endogenous PP2A inhibitors may be selected from one or more of SET,
25 SETBP1, and CIP2A (as described in Grech et al supra), preferably comprising CIP2A. Figure 2A shows significant upregulation of CIP2A in cell lines sensitive to a PP2A activator. The cancer may have an underexpression of one or more PP2A subunits. The PP2A subunits may be selected from one or more of PP2AA, PPP2R2A, PPPR2R5A, PPP2R5C and PP2Ac (as described in Grech et al supra).

30

Expression level

The overexpression or underexpression of a given marker in the cancer is typically determined by comparison to the level of the marker in normal cells of the same tissue type. The expression is thus typically normalized against the expression level of other genes, preferably comprising one or more housekeeping genes. A marker may also be classified as showing an overexpression or underexpression in a threshold percentage of a population of cancer patients. The overexpression or underexpression in each patient in the population may be higher than 2 from the geometric mean. At least 10%, more preferably at least 15% or more of the patients in the population may display such an overexpression or underexpression. The inventors have identified that such an overexpression of AURKA is present in at least 15% of stomach, colorectal, breast and pancreatic cancers (FIGURE 4).

Where the marker is overexpressed, its amount may be increased by any amount. For instance, the amount of AURKA or KIF2C may be increased in the cancer or tumour by at least 10%, at least 30% at least 40%, at least 50%, at least 60%, at least 70%, at least 80%, at least 90% or at least 95% compared with the level of AURKA or KIF2C in normal cells of the same type. The amount of both AURKA and KIF2C may each be increased in the cancer or tumour by at least 10%, at least 30% at least 40%, at least 50%, at least 60%, at least 70%, at least 80%, at least 90% or at least 95% compared with the levels of AURKA and KIF2C in normal cells of the same type.

Similarly, where the marker is underexpressed, its amount may be decreased by any amount, including to an undetectable amount or zero. For example, the amount of FIER2 and/or ER and/or PR may be increased in the cancer or tumour by at least 10%, at least 30% at least 40%, at least 50%, at least 60%, at least 70%, at least 80%, at least 90% or at least 95%) compared with the level(s) of the same marker(s) in normal cells of the same type.

The amount may be the amount of mRNA. The cancer may comprise an overexpression of AURKA and/or KIF2C mRNA. The cancer may comprise an increased amount of AURKA and/or KIF2C mRNA compared with normal cells of the same tissue type. The mRNA may be increased by any amount and in particular the % amounts discussed above. Similarly, a marker may be underexpressed in the cancer as described

above, with the cancer comprising an underexpression of mRNA of the marker in any amount, including in the % amounts discussed above. The amount of mRNA can be measured using a quantitative reverse transcription polymerase chain reaction (qRT-PCR), such as real time qRT-PCR, quantigene assay (Affymetrix/Thermo Fisher), by northern
5 blotting or using microarrays.

mRNA expression is preferably determined by comparing the gene expression of a sample to the distribution of expression levels of the specific gene across a reference sample composed of tumours that are diploid for that gene. A z-score may be derived using RNAseq by expectation maximisation (RSEM) algorithm (cBioportal for Cancer
10 Genomics, www.cbioportal.org; Gao et al, 2013 and Serami eta al 2012). A z-score of 2 SD higher or lower than the mean of the reference set is preferably considered as overexpression or underexpression respectively.

The amount may be the amount of protein. The cancer may comprise an overexpression of AURKA and/or KIF2C protein, such as compared with normal cells of
15 the same tissue type. The protein may be increased by any amount, including the % amounts discussed above. Where a marker is underexpressed in the cancer, it may be underexpressed at the protein level, with any decrease in protein amount, including in the % amounts discussed above. The amount of protein can be measured using immunohistochemistry, western blotting, mass spectrometry or fluorescence-activated cell
20 sorting (FACS). The thresholds for determining expression may vary between techniques used, and may be validated against immunohistochemistry scores.

The method of treating cancer in a patient may thus comprise (a) measuring the amount of AURKA and/or KIF2C in the cancer and (b) if the cancer comprises an overexpression of AURKA and/or KIF2C, administering to the patient a PP2A activator
25 and thereby treating the cancer. The amount of AURKA and/or KIF2C may be the mRNA or protein amount, and the overexpression any overexpression discussed above.

Sample

The above measurements may be carried out in any suitable sample from the
30 patient. The measurements may be carried out in a cancer or tumour biopsy obtained from the patient. The biopsy tissue may be formalin fixed paraffin embedded (FFPE) tissue or

fresh tissue. The tissue may be breast tissue, stomach tissue, colorectal tissue or pancreatic tissue. Any of the methods discussed above may be carried out on the cancer biopsy. Such methods may also be carried out on cancer cells circulating in the blood of the patient. The RNA methods may be carried out on urinary or blood exosomes.

Patient

Any patient may be treated in accordance with the invention. The patient is typically human. However, patient may be another mammalian animal, such as a commercially farmed animal, such as a horse, a cow, a sheep, a fish, a chicken or a pig, a laboratory animal, such as a mouse or a rat, or a pet, such as a guinea pig, a hamster, a rabbit, a cat or a dog.

Therapeutic agents

PP2A Activator

A PP2A activator is any molecule that increases PP2A function. The activator may increase PP2A function by any amount. The PP2A activator may increase PP2A function or activity by at least 10%, at least 30% at least 40%, at least 50%, at least 60%, at least 70%, at least 80%, at least 90% or at least 95%. The extent to which an activator increases PP2A function may be determined by measuring PP2A function in cells in the presence and absence of the activator. The cells may be normal cells or cancer cells. The cells may be cancer cells as described above. They may preferably be breast cancer cells, such as TNBC cells.

The activator may affect PP2A function by any means. It may increase or decrease the activity or amount of any molecule affecting PP2A function directly or indirectly. For instance, it may increase the amount of one or more PP2A subunits, such as one or more PP2A subunits as described above. It may stabilise a PP2A complex. It may reduce degradation of one or more PP2A subunits. It may increase the amount of active PP2A complex. It may decrease the activity or amount of one or more molecules inhibiting PP2A function, such as one or more endogenous PP2A inhibitors as described above. It may increase the degradation of one or more such endogenous PP2A inhibitors at the mRNA or

protein level, or decrease their function by inhibitory modification. It may decrease the transcription of a molecule inhibiting PP2A function, such as an endogenous inhibitor of PP2A. It may disrupt DNA encoding a molecule inhibiting PP2A function, such as an endogenous inhibitor of PP2A, using an agent such as a zinc finger nuclease.

5 The activator may act by binding the active site of the PP2A complex or act allosterically by binding at a different site. The activator may act by binding a regulator or ligand for the PP2A complex, to thereby enhance activation of PP2A or relieve inhibition of PP2A indirectly. The activator may be reversible or irreversible.

The PP2A activator may be a small molecule inhibitor, a peptide, a protein, an 10 antibody, a polynucleotide, an oligonucleotide, an antisense RNA, small interfering RNA (siRNA) or small hairpin RNA (shRNA).

Where the PP2A activator is a polynucleotide, it may encode any molecule increasing PP2A function. It may encode a PP2A subunit or any protein stabilising or activating a PP2A complex. It may encode a wildtype PP2A subunit, for example where a 15 cancer represents a mutated version thereof unable to provide for PP2A activity or unable to assemble a PP2A complex. Alternatively, the polynucleotide may act to reduce the amount of a molecule inhibiting PP2A function or acting to deregulate PP2A activity.

A polynucleotide, such as a nucleic acid, is a polymer comprising two or more nucleotides. The nucleotides can be naturally occurring or artificial. A nucleotide typically 20 contains a nucleobase, a sugar and at least one linking group, such as a phosphate, 2'O - methyl, 2' methoxy-ethyl, phosphoramidate, methylphosphonate or phosphorothioate group. The nucleobase is typically heterocyclic. Nucleobases include, but are not limited to, purines and pyrimidines and more specifically adenine (A), guanine (G), thymine (T), uracil (U) and cytosine (C). The sugar is typically a pentose sugar. Nucleotide sugars 25 include, but are not limited to, ribose and deoxyribose. The nucleotide is typically a ribonucleotide or deoxyribonucleotide. The nucleotide typically contains a monophosphate, diphosphate or triphosphate. Phosphates may be attached on the 5' or 3' side of a nucleotide.

Nucleotides include, but are not limited to, adenosine monophosphate (AMP), 30 adenosine diphosphate (ADP), adenosine triphosphate (ATP), guanosine monophosphate (GMP), guanosine diphosphate (GDP), guanosine triphosphate (GTP), thymidine

monophosphate (TMP), thymidine diphosphate (TDP), thymidine triphosphate (TTP), uridine monophosphate (UMP), uridine diphosphate (HDP), uridine triphosphate (UTP), cytidine monophosphate (CMP), cytidine diphosphate (CDP), cytidine triphosphate (CTP), 5-methylcytidine monophosphate, 5-methylcytidine diphosphate, 5-methylcytidine
5 triphosphate, 5-hydroxymethylcytidine monophosphate, 5-hydroxymethylcytidine diphosphate, 5-hydroxymethylcytidine triphosphate, cyclic adenosine monophosphate (cAMP), cyclic guanosine monophosphate (cGMP), deoxyadenosine monophosphate (dAMP), deoxyadenosine diphosphate (dADP), deoxyadenosine triphosphate (dATP), deoxyguanosine monophosphate (dGMP), deoxyguanosine diphosphate (dGDP),
10 deoxyguanosine triphosphate (dGTP), deoxythymidine monophosphate (dTMP), deoxythymidine diphosphate (dTDP), deoxythymidine triphosphate (dTTP), deoxyuridine monophosphate (dUMP), deoxyuridine diphosphate (dUDP), deoxyuridine triphosphate (dUTP), deoxycytidine monophosphate (dCMP), deoxycytidine diphosphate (dCDP) and deoxycytidine triphosphate (dCTP), 5-methyl-2' -deoxycytidine monophosphate, 5-methyl-
15 2'-deoxycytidine diphosphate, 5-methyl-2' -deoxycytidine triphosphate, 5-hydroxymethyl-2'-deoxycytidine monophosphate, 5-hydroxymethyl-2'-deoxycytidine diphosphate and 5-hydroxymethyl-2' -deoxycytidine triphosphate. The nucleotides are preferably selected from AMP, TMP, GMP, UMP, dAMP, dTMP, dGMP or dCMP.

The nucleotides may contain additional modifications. In particular, suitable
20 modified nucleotides include, but are not limited to, 2'amino pyrimidines (such as 2'-amino cytidine and 2'-amino uridine), 2'-hyrdroxyl purines (such as , 2'-fluoro pyrimidines (such as 2'-fluorocytidine and 2'fluoro uridine), hydroxyl pyrimidines (such as 5'-a-P-borano uridine), 2'-0-methyl nucleotides (such as 2'-0-methyl adenosine, 2'-0-methyl guanosine, 2'-0-methyl cytidine and 2'-0-methyl uridine), 4'-thio pyrimidines
25 (such as 4'-thio uridine and 4'-thio cytidine) and nucleotides have modifications of the nucleobase (such as 5-pentynyl-2'-deoxy uridine, 5-(3-aminopropyl)-uridine and 1,6-diaminohexyl-N-5-carbamoylmethyl uridine).

The nucleotides in the polynucleotide may be attached to each other in any manner. The nucleotides may be linked by phosphate, 2'0-methyl, 2' methoxy-ethyl,
30 phosphoramidate, methylphosphonate or phosphorothioate linkages. The nucleotides are typically attached by their sugar and phosphate groups as in nucleic acids. The nucleotides

may be connected via their nucleobases as in pyrimidine dimers.

The polynucleotide can be a nucleic acid, such as deoxyribonucleic acid (DNA) or a ribonucleic acid (RNA). The polynucleotide may be any synthetic nucleic acid known in the art, such as peptide nucleic acid (PNA), glycerol nucleic acid (GNA), threose nucleic acid (TNA), locked nucleic acid (LNA), morpholino nucleic acid or other synthetic polymers with nucleotide side chains. The polynucleotide may be single stranded or double stranded.

The polynucleotide sequence may be cloned into any suitable expression vector. In an expression vector, the polynucleotide sequence encoding a construct is typically operably linked to a control sequence which is capable of providing for the expression of the coding sequence by the host cell. Such expression vectors can be used to express a construct.

The term *"operably linked"* refers to a juxtaposition wherein the components described are in a relationship permitting them to function in their intended manner. A control sequence *"operably linked"* to a coding sequence is ligated in such a way that expression of the coding sequence is achieved under conditions compatible with the control sequences. Multiple copies of the same or different polynucleotide may be introduced into the vector.

The expression vector may then be introduced into a suitable host cell. Thus, a construct can be produced by inserting a polynucleotide sequence encoding a construct into an expression vector, introducing the vector into a compatible bacterial host cell, and growing the host cell under conditions which bring about expression of the polynucleotide sequence.

PP2A activator polynucleotides may reduce amounts of molecules acting to deregulate PP2A activity, such as endogenous PP2A inhibitors, present in the patient or the cancer, for example by knocking down their expression. Antisense and RNA interference (RNAi) technology for knocking down protein expression are well known in the art and standard methods can be employed to knock down expression of a molecule of interest. Both antisense and siRNA technology interfere with mRNA. Antisense oligonucleotides interfere with mRNA by binding to (hybridising with) a section of the mRNA. The antisense oligonucleotide is therefore designed to be complementary to the mRNA

(although the oligonucleotide does not have to be 100% complementary as discussed below). In other words, the antisense oligonucleotide may be a section of the cDNA. Again, the oligonucleotide sequence may not be 100% identical to the cDNA sequence. This is also discussed below. RNAi involves the use of double-stranded RNA, such small interfering RNA (siRNA) or small hairpin RNA (shRNA), which can bind to the mRNA and inhibit protein expression.

Accordingly, the activator may comprise an oligonucleotide which specifically hybridises to an mRNA encoding an inhibitor of PP2A function, such as an endogenous PP2A inhibitor described above. An oligonucleotide *"specifically hybridises"* to a target sequence when it hybridises with preferential or high affinity to the target sequence but does not substantially hybridise, does not hybridise or hybridises with only low affinity to other sequences. More preferably, the oligonucleotide hybridises to the target sequence with a T_m that is at least 5 °C, at least at least 10 °C, at least 20 °C, at least 30 °C or at least 40 °C, greater than its T_m for other nucleic acids. Conditions that permit the hybridisation are well-known in the art (for example, Sambrook et al., 2001, Molecular Cloning: a laboratory manual, 3rd edition, Cold Spring Harbour Laboratory Press; and Current Protocols in Molecular Biology, Chapter 2, Ausubel et al., Eds., Greene Publishing and Wiley-lnterscience, New York (1995)). The hybridisation conditions may be stringent conditions as described in the art.

Oligonucleotides are short nucleotide polymers which typically have 50 or fewer nucleotides, such 40 or fewer, 30 or fewer, 22 or fewer, 21 or fewer, 20 or fewer, 10 or fewer or 5 or fewer nucleotides. The oligonucleotide used may be 20 to 25 nucleotides in length, more preferably 21 or 22 nucleotides in length. The nucleotides can be naturally occurring or artificial. The nucleotides can be any of those described above.

The PP2A activator may be an antibody which specifically binds to any target molecule (typically a protein) so as to increase PP2A function directly or indirectly.

The target molecule may be a component of the PP2A complex. In this aspect, the antibody may bind to the component in such a way that activates PP2A function, including by displacing or blocking interaction between the component and a PP2A inhibitor, or by allosteric activation of the PP2A complex. Alternatively, the target molecule may be an inhibitory ligand for the PP2A complex or any other PP2A inhibitor or molecule acting to

deregulate PP2A activity. In this aspect, the antibody typically binds to the target molecule to antagonise its activity, for example by blocking its binding to the PP2A complex or sequestering the target molecule. An antagonist antibody may act to decrease function of an endogenous PP2A inhibitor as described above.

An antibody "specifically binds" to a protein when it binds with preferential or high affinity to that protein but does not substantially bind, does not bind or binds with only low affinity to other proteins. For instance, an antibody "specifically binds" a target molecule when it binds with preferential or high affinity to that target but does not substantially bind, does not bind or binds with only low affinity to other human proteins.

An antibody binds with preferential or high affinity if it binds with a Kd of 1 x 10-7 M or less, more preferably 5 x 10-8 M or less, more preferably 1 x 10-8 M or less or more preferably 5 x 10-9 M or less. An antibody binds with low affinity if it binds with a Kd of 1 x 10-6 M or more, more preferably 1 x 10-5 M or more, more preferably 1 x 10-4 M or more, more preferably 1 x 10-3 M or more, even more preferably 1 x 10-2 M or more.

The antibody may be, for example, a monoclonal antibody, a polyclonal antibody, a single chain antibody, a chimeric antibody, a bispecific antibody, a CDR-grafted antibody or a humanized antibody. The antibody may be an intact immunoglobulin molecule or a fragment thereof such as a Fab, $F(ab')_2$ or Fv fragment.

Preferred PP2A activators include small molecule inhibitors of endogenous PP2A inhibitors or oligonucleotides reducing expression of these as described above. The endogenous PP2A inhibitor may be IGBP1, SET, SETBP1 or CIP2A.

APP2A activator may increase PP2A-mediated dephosphorylation of c-Myc. A PP2A activator may reduce inhibition of PP2A-mediated dephosphorylation of c-Myc by CIP2A. The PP2A activator may be a CIP2A inhibitor. It may downregulate CIP2A mRNA. It may direct CIP2A for proteasome-dependent degradation. Examples of compounds acting to inhibit CIP2A include bortezomib, celastrol and ethoxysanguinaire and functional derivatives and analogues thereof.

The PP2A activator may be a SET inhibitor. A SET inhibitor may be an ApoE mimetic peptide binding to SET to inhibit its binding to PP2A. The SET inhibitor may disrupt a SET/PP2A complex. A preferred SET inhibitor is FTY720 (Fingolimod) or a functional derivative or analogue thereof, or any other analogue of myriocin acting as a

PP2A activator. An example of an FTY720 analogue is OSU-2S. Another compound that may be used as a SET inhibitor is ceramide or any agent stimulating ceramide biosynthesis, such as vitamin D3. An agent stimulating ceramide biosynthesis may be used in combination with ceramide.

5 The PP2A activator may be a SETBP1 inhibitor. It may prevent protection of SET from proteolytic cleavage by disrupting binding between SET and SETBP1.

The PP2A activator may be an IGBP1 inhibitor. It may prevent binding of IGBP1 to the catalytic subunit of PP2A.

APP2A activator may inhibit the phosphorylation of the catalytic subunit of PP2A 10 and/or the demethylation of the C-terminus of the catalytic subunit of PP2A.

Molecules upregulating expression of PP2A subunits may also be used as PP2A activators. An example includes methylprednisole or a functional derivative thereof. Other molecules that may be used as activators of PP2Aby a variety of mechanisms include endostatin, forskolin, dithiolethionines such as anethole dithiolethione (ADT), carnosic 15 acid, and functional derivatives and analogues thereof. Small molecule activators of PP2A are also described in McClinch et al (Therapeutic reactivation of PP2Afor prostate cancer treatment; 2015 Nov 5-9, Molecular Targets and Cancer Therapeutics, Boston).

Further information on provision of PP2A inhibitors is provided in Grech et al, supra, incorporated by reference herein.

20 The method of treatment with a PP2A activator may further comprise administration of an AURKA and/or KIF2C antagonist as described below.

AURKA and KIF2C antagonists

An AURKA antagonist is any molecule that decreases AURKA function. Similarly, 25 a KIF2C antagonist is any molecule that decreases KIF2C function. An antagonist may decrease both AURKA and KIF2C function. The function may be decreased in any % amount discussed above by reference to PP2A function. The antagonist of AURKA or KIF2C may increase PP2A activity in any % amount discussed above. The extent to which an antagonist decreases function may be determined by measuring AURKA or KIF2C 30 function in cells in the presence and absence of the antagonist. The cells may be normal

cells or cancer cells. The cells may be cancer cells as described above. They may preferably be breast cancer cells, such as TNBC cells.

The antagonist may decrease AURKA or KIF2C function by any means. It may increase or decrease the activity or amount of any molecule affecting AURKA or KIF2C function directly or indirectly. It may decrease the amount of AURKA and/or KIF2C at the mRNA or protein level. It may increase degradation of AURKA and/or KIF2C. It may decrease the function of AURKA and/or KIF2C by inhibitory modification. It may decrease the transcription of a molecule enhancing AURKA or KIF2C function. It may disrupt DNA encoding AURKA or KIF2C or a molecule enhancing AURKA or KIF2C function, using an agent such as a zinc finger nuclease.

The antagonist may act by binding the active site of AURKA or act allosterically by binding at a different site. The antagonist may act by binding a regulator or ligand for AURKA or KIF2C, to thereby reduce activation of AURKA or KIF2C. The antagonist may be reversible or irreversible.

An AURKA antagonist or a KIF2C antagonist may be a small molecule inhibitor, a peptide, a protein, an antibody, a polynucleotide, an oligonucleotide, an antisense RNA, small interfering RNA (siRNA) or small hairpin RNA (shRNA). Selection of molecules such as antibodies, polynucleotides, oligonucleotides for targeting a gene of interest is described in detail above in relation to PP2A activators. The same criteria are described herein for selection of antagonists of AURKA and KIF2C.

An antagonist of AURKA or KIF2C may be an oligonucleotide which specifically hybridises to an mRNA encoding AURKA or KIF2C or an mRNA encoding a molecule which enhances AURKA or KIF2C activity. An antagonist of AURKA or KIF2C may be a polynucleotide encoding any molecule that decreases AURKA or KIF2C function. An antagonist of AURKA or KIF2C may be an antibody which specifically binds to any target molecule (typically a protein) so as to decrease AURKA or KIF2C function directly or indirectly. The antagonist may be an antibody specifically binding AURKA or KIF2C. In this aspect, the antibody may decrease AURKA or KIF2C function by allosteric inactivation or by blocking interaction between its target and a ligand required for activity. An antagonist antibody specifically binding AURKA or KIF2C may decrease AURKA or KIF2C function in any % amount discussed above with reference to PP2A function.

Pharmaceutical Compositions and Modes of Administration

The agents for use in the methods of treatment described herein may be formulated in pharmaceutical compositions. These compositions may comprise, in addition to the therapeutically active ingredient(s), a pharmaceutically acceptable excipient, carrier, diluent, buffer, stabiliser or other materials well known to those skilled in the art. Such materials should be non-toxic and should not interfere with the efficacy of the active ingredient. The pharmaceutical carrier or diluent may be, for example, an isotonic solution.

The precise nature of the carrier or other material may depend on the route of administration, e.g. oral, intravenous, cutaneous or subcutaneous, nasal, intramuscular and intraperitoneal routes. Examples of suitable compositions and methods of administration are provided in Esseku and Adeyeye (2011) and Van den Mooter G. (2006). For example, solid oral forms may contain, together with the active substance, diluents, e.g. lactose, dextrose, saccharose, cellulose, corn starch or potato starch; lubricants, e.g. silica, talc, stearic acid, magnesium or calcium stearate, and/or polyethylene glycols; binding agents; e.g. starches, gum arabic, gelatin, methylcellulose, carboxymethylcellulose or polyvinyl pyrrolidone; disaggregating agents, e.g. starch, alginic acid, alginates or sodium starch glycolate; effervescing mixtures; dyestuffs; sweeteners; wetting agents, such as lecithin, polysorbates, laurylsulphates; and, in general, non-toxic and pharmacologically inactive substances used in pharmaceutical formulations. Such pharmaceutical preparations may be manufactured in known manner, for example, by means of mixing, granulating, tabletting, sugar-coating, or film-coating processes.

Oral formulations include such normally employed excipients as, for example, pharmaceutical grades of mannitol, lactose, starch, magnesium stearate, sodium saccharine, cellulose, magnesium carbonate, and the like. These compositions take the form of solutions, suspensions, tablets, pills, capsules, sustained release formulations or powders and contain 10% to 95% of active ingredient, preferably 25% to 70%. Where the pharmaceutical composition is lyophilised, the lyophilised material may be reconstituted prior to administration, e.g. a suspension. Reconstitution is preferably effected in buffer.

Capsules, tablets and pills for oral administration to an individual may be provided with an enteric coating comprising, for example, Eudragit "S", Eudragit "L", cellulose acetate, cellulose acetate phthalate or hydroxypropylmethyl cellulose.

Liquid dispersions for oral administration may be syrups, emulsions or suspensions. 5 The syrups may contain as carriers, for example, saccharose or saccharose with glycerine and/or mannitol and/or sorbitol.

Suspensions and emulsions may contain as carrier, for example a natural gum, agar, sodium alginate, pectin, methylcellulose, carboxymethylcellulose, or polyvinyl alcohol. The suspensions or solutions for intramuscular injections may contain, together with the 10 active substance, a pharmaceutically acceptable carrier, e.g. sterile water, olive oil, ethyl oleate, glycols, e.g. propylene glycol, and if desired, a suitable amount of lidocaine hydrochloride.

Solutions for intravenous administration or infusion may contain as carrier, for example, sterile water or preferably they may be in the form of sterile, aqueous, isotonic 15 saline solutions.

For suppositories, traditional binders and carriers may include, for example, polyalkylene glycols or triglycerides; such suppositories may be formed from mixtures containing the active ingredient in the range of 0.5% to 10%, preferably 1% to 2%.

Polynucleotide or oligonucleotide inhibitors maybe naked nucleotide sequences or be 20 in combination with cationic lipids, polymers or targeting systems. They may be delivered by any available technique. For example, the polynucleotide or oligonucleotide may be introduced by needle injection, preferably intradermally, subcutaneously or intramuscularly. Alternatively, the polynucleotide or oligonucleotide may be delivered directly across the skin using a delivery device such as particle-mediated gene delivery. 25 The polynucleotide or oligonucleotide may be administered topically to the skin, or to mucosal surfaces for example by intranasal, oral, or intrarectal administration.

Uptake of polynucleotide or oligonucleotide constructs may be enhanced by several known transfection techniques, for example those including the use of transfection agents. Examples of these agents include cationic agents, for example, calcium phosphate and 30 DEAE-Dextran and lipofectants, for example, lipofectam and transfectam. The dosage of the polynucleotide or oligonucleotide to be administered can be altered.

Administration is typically in a "prophylactically effective amount" or a "therapeutically effective amount" (as the case may be, although prophylaxis may be considered therapy), this being sufficient to show benefit to the individual, e.g. an effective amount to prevent or delay onset of the disease or condition, to ameliorate one or more
5 symptoms, to induce or prolong remission, or to delay relapse or recurrence.

The dose may be determined according to various parameters, especially according to the substance used; the age, weight and condition of the individual to be treated; the route of administration; and the required regimen. A physician will be able to determine the required route of administration and dosage for any particular individual. Atypical
10 daily dose is from about 0.1 to 50 mg per kg of body weight dependent on the conditions mentioned above. The dose may be provided as a single dose or may be provided as multiple doses, for example taken at regular intervals, for example 2, 3 or 4 doses administered hourly. Typically polynucleotide or oligonucleotide inhibitors are administered in the range of 1 pg to 1 mg, preferably to 1 pg to 10 μg nucleic acid for
15 particle mediated delivery and 10 μg to 1 mg for other routes.

Examples of the techniques and protocols mentioned above can be found in Remington's Pharmaceutical Sciences, 20th Edition, 2000, pub. Lippincott, Williams & Wilkins.

A composition may be administered alone or in combination with other therapeutic
20 compositions or treatments, for example as adjunct therapy. The other therapeutic compositions or treatments may for example be one or more of those discussed herein, and may be administered either simultaneously or sequentially with the composition or treatment of the invention. Thus, the PP2A activator may be administered in combination with any other cancer therapy. The other cancer therapy may be selected from any known
25 therapy for the relevant cancer. The other cancer therapy may comprise targeting of a molecule such as HER2, ER or PR. In particular where HER2, ER or PR is overexpressed in the cancer. The cancer therapy may comprise administration of an antagonist of HER2, ER or PR (or antagonists for more than one of HER2, ER and PR). These antagonists may be a small molecule inhibitor, a peptide, a protein, an antibody, a polynucleotide, an
30 oligonucleotide, an antisense RNA, small interfering RNA (siRNA) or small hairpin RNA (shRNA), selected according to criteria discussed above in relation to PP2A activators,

applied to selection of molecules affecting HER21, ER or PR activity. Such molecules may antagonise the activity of HER2, ER or PR such as by decreasing their expression or specifically binding their encoding mRNA. The antagonists may be selected from antibodies specifically binding HER, ER or PR.

5

Medical Uses

The invention also provides medical uses corresponding to the methods of treatment described above. The invention thus provides a PP2A activator for use in treating cancer in a patient, wherein the cancer comprises an overexpression of AURKA and/or
10 KIF2C. It also provides use of a PP2A activator in the manufacture of a medicament for treating cancer in a patient, wherein the cancer comprises an overexpression of AURKA and/or KIF2C. The cancer and PP2A activator may be selected from any described above in relation to methods of treatment.

15 *Selection of patients*

A patient may be treated with a PP2A activator as described above without previous determination that the patient overexpresses AURKA or KIF2C. This may occur where the patient is suspected of having deregulated PP2A activity or shows resistance to other
20 treatments indicating that an overexpression of AURKA or KIF2C is likely to be present. Alternatively, a patient may be selected for treatments described above on the basis of being known to have a cancer comprising an overexpression of AURKA and/or KIF2C. The method of treatment may thus comprise a step of measuring expression of AURKA and/or KIF2C as described above. An overexpression of AURKA and/or KIF2C may be
25 determined as described above.

The patient may also be selected for said treatment on the basis of the cancer having an underexpression of one or more of HER2, ER and PR, as described above. The cancer may have an underexpression of each of HER2, ER and PR. The patient may thus be predicted to be resistant to therapy antagonising one or more of (or all of) HER2, ER
30 and PR, such that therapy with a PP2A activator is appropriate. Such a patient may further

be selected for said treatment based on having an overexpression of AURKA and/or KIF2C.

The selection of the patient for treatment may be carried out on diagnosis of the cancer, during treatment of the cancer, or following resistance to a cancer therapy. The 5 cancer therapy to which resistance is shown may comprise antagonism of one or more of HER2, ER and PR. Where selection is carried out on diagnosis, the diagnosis may be made by any means, and based on any symptoms. Selection may also be made based on a predisposition of the patient for a particular cancer, or a risk of the patient having a particular cancer.

10 In a related aspect, the invention provides a kit for treating cancer comprising (a) one or more reagents suitable for measuring expression of AURKA and/or KIF2C and (b) a PP2A activator. The kit thus provides components suitable both for determining whether a cancer has an overexpression of markers correlating with responsiveness to a PP2A activator, and a therapeutic agent for treatment purposes, the 15 PP2A activator. Reagents suitable for measuring expression of AURKA and/or KIF2C are discussed in more detail below.

The kit may additionally comprise means for the measurement of other laboratory or clinical parameters. The kit may additionally comprise one or more other reagents or instruments which enable the method to be carried out. Such reagents or instruments 20 include one or more of the following: suitable buffer(s) (aqueous solutions), means to isolate AURKA and/or KIF2C from a sample, means to obtain a sample from the individual (such as a vessel or an instrument comprising a needle) or a support comprising wells on which quantitative reactions can be done.

The kit may comprise instructions for determining whether there is an 25 overexpression of AURKA and/or KIF2C in the cancer, typically in a sample from a patient, as described above. The kit may comprise details regarding which individuals the method may be carried out upon. The individuals may be patients having the criteria for selection for treatment described above.

Another kit of the invention for treating cancer comprises a PP2A activator and said 30 details regarding which individuals the method may be carried out upon and/or said instructions for determining whether there is an overexpression of AURKA and/or KIF2C

in a cancer. The reagents suitable for measuring expression of AURKA and/or KIF2C may then be provided separately to the kit.

Detection/diagnosis/prognosis

5

Methods

Based on the correlation between overexpression of AURKA and/or KIF2C and cancer, including particular subtypes of cancer, the invention further provides uses of AURKA and/or KIF2C in classification of cancer, and corresponding methods and
10 systems. The invention further provides for prediction of responsiveness to treatment. The invention also provides for diagnosis and prognosis of cancer. Diagnosis includes determining whether or not an individual has a cancer or tumour and/or determining the severity of the cancer or tumour. Prognosis includes predicting whether or not an individual will develop a cancer or tumour, whether or not they will need treatment, the
15 type of treatment the individual will need, whether or not they will respond to a treatment, whether or not and/or when they will suffer a cancer episode, recurrence or relapse, and the severity or duration of a symptom or a cancer episode, recurrence or relapse.

The invention thus provides a method for detecting or diagnosing a cancer sensitive to a PP2A activator, in a patient, the method comprising measuring expression of AURKA
20 and/or KIF2C in the patient, wherein overexpression of AURKA and/or KIF2C indicates that the patient comprises said cancer.

Further described herein is a method for prognosing a cancer in a patient, the method comprising determining whether or not the cancer comprises an overexpression of AURKA and/or KIF2C, wherein an overexpression of AURKA and/or KIF2C in the
25 cancer indicates that the patient has a worse prognosis than in the situation of normal expression of AURKA and/or KIF2C. The method of prognosis may predict whether or not an individual in remission from cancer will have a recurrence. Predicting whether or not the individual will have a recurrence includes determining the likelihood that the individual will have a recurrence, and/or predicting when they will have a recurrence. In
30 some cases the individual may be in remission following treatment with a therapy comprising antagonism of HER2 and/or ER and/or PR as described herein.

The invention additionally provides a method for determining whether or not a patient having or suspected of having or being at risk of developing cancer will respond to treatment with a PP2A activator, which method comprises measuring expression of AURKA and/or KIF2C in the individual, and thereby predicting whether or not the patient will respond to treatment with a PP2A activator. Additionally to predicting responsiveness to treatment with a PP2A activator, the method may also further predicting responsiveness to treatment with an AURKA antagonist and/or a KIF2C antagonist.

Predicted responsiveness in an individual to a given therapy means that the individual is expected to derive benefit, or a sufficient extent of benefit, from receiving the therapy. Predicted non-responsiveness in an individual to a therapy means that the individual is not expected to derive benefit, or a sufficient extent of benefit, from receiving the therapy. The method for predicting the response may be carried out before administration of the PP2A activator. The PP2A activator may be administered with an AURKA antagonist and/or KIF2C antagonist. The prediction may then be taken into account when selecting or recommending a suitable treatment for the individual. Alternatively, the method may be carried out after treatment with the therapy and used to monitor and predict the individual's response to treatment. Typically the method is for predicting whether or not the individual will have a primary response to treatment with the therapy, i.e. whether or not the individual will respond when first receiving the treatment. In some cases the method is for predieting secondary non-responsiveness, i.e. whether or not an individual who initially responds to treatment will later stop responding to treatment or will respond less well to the treatment.

According to the present invention, an increased level of AURKA and/or KIF2C in an individual, as compared with a reference sample or reference level, indicates a positive diagnosis relating to the presence of cancer, for example that the individual has a cancer or a particular form of cancer or has more severe cancer. An increased level of AURKA and/or KIF2C also indicates a negative prognosis, that is a poor predicted outcome for the individual, for example that the individual will not respond to a particular therapy, that an individual in remission from cancer will have a recurrence or that the individual is at increased risk of developing the cancer.

Conversely, a decreased or normal level of AURKA and/or KIF2C indicates a negative diagnosis, for example that the individual does not have the cancer or has less severe cancer. A decreased level of AURKA and/or KIF2C may indicate a positive prognosis, that is a good outcome for the patient, for example that the individual will respond to a particular therapy or that an individual in remission from the cancer will not have a recurrence or is not at increased risk of developing the disease or condition. For diagnosing whether or not an individual has the cancer, the reference sample or level typically represents a baseline level of AURKA and/or KIF2C in an individual who does not have the relevant cancer, or who is suspected of having a cancer, but is subsequently confirmed to not have the cancer.

The method of diagnosis or prognosis may include selecting or recommending a suitable treatment for the individual, i.e. based on the diagnosis or prognosis. The selected or recommended treatment may then be administered to the individual. For example, an overexpression of AURKA and/or KIF2C, as compared with a reference sample or reference level, indicates that the individual will respond to therapy with a PP2A activator. A therapy including use of a PP2A activator may then be selected or recommended, and may then further be administered to the individual. Similarly, a therapy comprising use of an AURKA antagonist and/or KIF2C antagonist may be selected based on the overexpression of AURKA and/or KIF2C.

In other cases, a decreased or normal level of AURKA and/or KIF2C, as compared with a reference sample or reference level, indicates that the individual will not respond to therapy with a PP2A activator, and/or may not have a deregulation of PP2A. A PP2A activator therapy is then not administered to the individual. Further, a therapeutic treatment other than a PP2A activator may be selected or recommended for treatment of the individual, and may then further be administered to the individual.

In all aspects of the invention, an individual having cancer (e.g. breast cancer or basal breast cancer) or an individual suspected of having the disease or condition and/or an individual at risk of developing the disease or condition may be selected for treatment or identified. For example, the individual may not have been formally diagnosed but may be suspected of having the disease or condition because of the presence of one or more symptoms. The individual may be considered at risk of developing cancer if they have one

or more risk factors associated with cancer and/or one or more predispositions which increase their susceptibility to cancer.

The invention also provides a method for classifying a cancer in a patient, the method comprising measuring expression of AURKA and/or KIF2C in the patient, and classifying the cancer as of a particular subtype based on the expression. The method may comprise classifying the cancer as of a more aggressive subtype, based on overexpression of AURKA and/or KIF2C. In particular aspects, a breast cancer may be classified as a luminal or basal breast cancer, wherein overexpression of AURKA and/or KIF2C classifies the cancer as a basal breast cancer, and wherein normal or decreased expression of AURKA and/or KIF2C classifies the cancer as a luminal breast cancer. The methods for classifying a cancer may further comprising measuring expression of one or more of HER2, ER and PR to further classify the cancer as positive for expression or overexpression of one or more of such markers. Preferably expression of HER2 is further measured as part of the classification. The cancer may be classified as triple negative for expression of HER2, ER and PR, as showing decreased expression or no detectable expression of any of the three markers.

In all of the above methods, expression of both AURKA and KIF2C may be measured in the patient. An overexpression of AURKA and/or KIF2C may be determined as described above. AURKA and/or KIF2C are measured and compared with reference levels for normal AURKA and/or KIF2C expression as discussed above, typically in normal tissue of the same type as the cancer. The cancer may be any cancer or tumour described above, and is preferably stomach, colorectal, pancreatic, or breast cancer or tumour, most preferably a breast cancer or tumour. The cancer may be a basal breast cancer

All of the above methods may further comprise measuring expression of one or more of HER2, ER and PR in the patient, or any combination thereof as described above, including all of HER2, ER and PR. The methods may also further comprise determining whether there is a deregulation of PP2A in the patient by detection of expression of other markers of PP2A deregulation, or determination of reduced PP2A activity by any means discussed above. Markers of PP2A deregulation may comprise overexpression of one or more endogenous PP2A inhibitors, such as one or more of CIP2A, SET, IGBPB1 and

SETBPl. Markers of PP2A deregulation may comprise underexpression or mutation of one or more PP2A subunits as described above.

Kits

5 In related aspects, the invention provides a kit for detecting a cancer comprising reagents suitable for detecting expression of AURKA and/or KIF2C. The kit typically further comprises reagents for detecting expression of one or more other genes to allow for normalisation of AURKA/KIF2C levels, preferably reagents for detection of expression of one or more housekeeping genes. The one or more housekeeping genes allow for 10 normalisation of the expression detected, to more sensitively detect a relevant change in expression level of a marker to be used for detection of cancer. The kit may comprise reagents for detection of expression of at least one, two, three, four, five, or eight, or at least ten housekeeping genes. The kit may be for detecting a cancer comprising a deregulation of PP2A. The kit may be for detecting a cancer responsive to treatment with a 15 PP2A activator.

The kit preferably comprises reagents suitable for detecting expression of AURKA and KIF2C. The kit may further comprise reagents suitable for detecting expression of one or more endogenous inhibitors of PP2A or one or more PP2A subunits. Thus the kit may comprise reagents suitable for detecting expression of one or more of CIP2A, SET, 20 SETBP 1, IGBP 1, such as CIP2A and SET, SET and SETBP 1, CIP2A and SETBP 1, CIP2A and IGBPl, SET and IGBPl; and SETBPl and IGBPl, or of expression of all of CIP2A, SET, SETBP, IGBPl. The kit preferably comprises a reagent suitable for detecting expression of CIP2A. The kit may alternatively or additionally comprise reagents suitable for detecting expression of one or more of PP2AA, PPP2R2A, PPPR2R5A, PPP2R5C and 25 PP2Ac. The kit preferably comprises reagents suitable for detecting expression of PP2Ac and/or PPP2R2A.

In some embodiments, the only reagents for detecting expression of genes (at the protein, or mRNA level) included in the kit may be reagents detecting the combinations of genes listed above. Thus, for example the kit may comprise reagents for detecting 30 expression of AURKA and/or KIF2C, and one or more housekeeping genes, and no other reagents for detection of expression of any genes. Such a kit may comprise reagents for

detection of expression of up to two, up to five or up to ten housekeeping genes. Such a kit may include additional reagents for detection of expression of one or more endogenous PP2A inhibitors as described above, and/or for detection of expression of one or more PP2A inhibitors as described above, and no other reagents for detection of expression of any genes.

The reagents are typically suitable for measuring the expression level of the relevant marker. In this manner, whether the cancer displays an overexpression or underexpression of the marker may be determined by comparison with a reference level, as described above. The reagents may be suitable for measuring mRNA expression or protein expression. The reagents suitable for detecting expression of particular markers may be selected from nucleic acid probes or primers and antibodies specifically hybridising to the nucleic acid sequence of the relevant marker or specifically binding the marker protein.

Standard methods known in the art may be used to assay the level of a marker. mRNA levels can be accurately quantified by RNA analysis methods including Quantigene® assay (Affymetrix/ Thermo Fisher), qRT-PCR and next generation sequencing. Methods may alternatively involve using an agent that binds to or reacts with the relevant protein. The agent may be incubated with the sample from the individual and complex formation or a reaction between the agent and the relevant protein is measured. The agent typically binds specifically to the protein. The agent may be an antibody specific for the protein or an aptamer that binds to the protein.

A variety of protocols for competitive binding or immunoradiometric assays to determine the specific binding capability of compounds, such as antibodies or antibody constructs and oligonucleotides are well known in the art (see for example Maddox et al, J. Exp. Med. 158, 1211-1226, 1993). Antigen-capture dipstick assays and Enzyme-linked Immunosorbant Assay (ELISA) may be used. ELISA is typically carried out using the sandwich technique or the competitive technique, which are known to those in the art. The invention may also employ antibodies in direct sensing techniques including but not limited to those based upon surface plasmon resonance, surface acoustic wave, quartz crystal microbalance, microcalorimetry or electrochemical impedance spectroscopy. Expression levels may be determined by, for example, flow cytometry or by quantitative immunohistochemistry analysis on histological sections of patient intestinal tissue.

Systems

In related aspects the invention provides a system for classifying cancer in a patient, or for predicting responsiveness of a cancer patient to treatment with a PP2A activator, the system comprising:

(a) a measuring module for determining expression of AURKA and/or KIF2C in the patient,

(b) a storage module configured to store control data and output data from the measuring module,

(c) a computation module configured to provide a comparison between the value of the output data from the measuring module and the control data; and

(d) an output module configured to display whether or not the patient has cancer based on the comparison,

wherein an overexpression of AURKA and/or KIF2C in the patient classifies the cancer, or predicts that the patient will respond to treatment with a PP2A activator.

The measuring module may be configured for determining expression of one or more other markers for detection of cancer described herein, and the other modules configured to further take into account expression of such markers in the determination of the presence of cancer, classification of cancer, prognosis of cancer or prediction of responsiveness to treatment with a PP2A activator. The correlation between expression of the other markers and the detection, diagnosis or prognosis is made as described above.

EXAMPLES

Example 1 – Identification of markers of sensitivity to PP2A activators

Data analysis of well annotated, publicly available databases (TCGA) showed that PP2A deregulation is a common event in breast cancer, with 59.6% of basal breast cancer patients showing a low expression of one of the PP2A complex components or a high expression of the inhibitory subunits.

The main aim of the study was to identify genes whose expression depends directly on PP2A activity. In the ideal scenario, these novel biomarkers would reflect PP2A activity regardless of the mechanism driving the activity regulation.

We first established cell line models to investigate possible markers of sensitivity to PP2A activators. Basal breast cancer cell line MDAMB23 1 was sensitive to the PP2A activator, FTY720, and showed a suppressed PP2A activity while the MDAMB453 Luminal cell line was not sensitive to FTY720 and had relatively high PP2A activity. Drug sensitivity is the inverse of percent viability.

In more detail, the dose-dependent effect of FTY720 on breast cancer cell lines provided information on the sensitivity of the cell line to PP2A activation. Cytotoxicity results for the 12 breast cancer cell lines tested are illustrated in Figure 1 where the % cell viability is expressed as a percentage of the parallel control culture (without drug) against FTY720 concentration. The average vehicle control (VC) % cell viability for each cell line was never under 85% viability compared to the untreated ruling out any interference by the vehicle. The IOμM and the 25μM doses were considered to be cytotoxic as cellular morphology was generally altered, and hence are not shown.

The three sensitive cell lines, defined by % cell viability under 50% (IC50) at a dose lower than 5μM, were represented by a Triple Negative phenotype (Figure 1A). BT-20 was the most sensitive at 0.5μM, followed by MDAMB23 1 and Hs578T showing and IC50 at 2.5μM FTY720 and 5μM FTY720, respectively. From this experiment, the effective dose of FTY720, that is not lethal, was set at 5μM for assessing the effect of FTY720 on RNA expression. MCFlOAis a cell-line derived from normal epithelium. This cell line was used as a baseline control in the study of the PP2A mechanism in breast cancer cell lines.

Example 2 - PP2A biomarker discovery and evaluation

The knowledge gained from the cellular models implied the need to characterise novel biomarkers for PP2A activity. The PP2A deregulation criteria was designed based on this information to probe datasets for gene candidates.

Commonly deregulated regulatory subunits of PP2A were identified. Preliminary

analysis of the PP2A regulators was done on TCGA data to assess possible pathways for deregulation within breast cancer.

Initial analysis of transcript expression and protein expression of PP2A regulators in breast cancer cellular models did not identify clear markers that consistently reflect FTY720 sensitivity. Moreover, the assessment of PP2A regulators and phosphotargets of PP2A, in local patients and a set of tissue arrays (TMAs), highlighted the complexity and heterogeneity of the feedback mechanism. Biomarkers that directly measure the activity of the complex or the specific function of the variety of complexes were required to predict sensitivity to drugs at a tissue level.

A number of candidate PP2A biomarker genes were identified. As outlined in various studies, a biomarker needs to be well defined in its objectives, use and target population (Altar et al., 2007; Pepe, Feng, Janes, Bossuyt, & Potter, 2008). The approach for biomarker discovery involved the use of breast cancer data in the TCGA data portals in the context of PP2A deregulation. Genes were shortlisted using network analysis and known biological evidence connecting biomarkers to the PP2A pathway.

Following discovery, the viability of using these biomarkers in the same cohort based on RNASeqV2 expression was assessed. Validation analysis was performed using the TCGA RNASeqV2 data analysed manually providing a better insight into the data. The in silico analysis was then compared to observations within the cellular models and more importantly a cohort of local patients (N = 95 tumours, 3 Relapse tumours, 7 in situ, non-invasive tumours, 36 Normal breast tissue, of which 33 were patient-matched with tumours).

The set of biomarkers was validated and evidence-based selection of biomarker candidates, identified AURKA and KTF2C as strong predictors of sensitivity to FTY720. These genes were used to classify patients and cell lines into those that are eligible to PP2A activation and those that are not predicted to have PP2A deregulation. The novel therapeutic class was mainly represented by the Basal or Triple Negative groups and was associated with higher tumour aggressivity.

In more detail, the expression of the PP2A complex and regulator genes was measured in the 12 breast cancer cell lines when untreated, but also when treated with FTY720 (5μM).

Figure 2 shows that Cip2a (KIAA1524) RNA expression correlates with sensitivity of breast cancer cell lines to FTY720 **(Figure 2).** MDA-MB-23 1 exemplifies FTY720-sensitive breast cancer cell lines, showing a low PP2A enzymatic activity and a high Cip2a RNA expression relative to MCF10A (a cell line derived from normal epithelium).

5 AURKA and KIF2C RNA expression were found to be positively correlated with FTY720-sensitive breast cancer cell lines **(Figure 2B).** Of interest expression also correlated positively with PP2A endogenous inhibitors, cip2a and SET **(Table 1).**

Table 1: Spearman correlation analysis between the expression of the PP2A complex subunits and the 2 biomarkers.

The normalised RNASeqV2 data as downloaded from the TCGA database was used.

[N=873 tumour cases; Sig. (2-tailed) stands for two-tailed significance, significance at the 95% confidence interval is highlighted]

		AURKA	**KIF2C**
PPP2CA	Spearman Correlation	-0.056	-0.181
	Sig. (2-tailed)	0.107	< 0.001*
PPP2R2A	Spearman Correlation	-0.262	-0.209
	Sig. (2-tailed)	< 0.001*	0.005*
CIP2A	Spearman Correlation	0.750	0.773
	Sig. (2-tailed)	< 0.001*	< 0.001*

10

In addition, the expression of the genes within breast cancer tissue was also compared to the respective patient matched control tissue **(Figure 3B).** AURKA and KXF2C RNA expression was found to be significantly higher in tumour material relative to matched normal tissues. This is also seen when patients are subdivided by the hormone

15 status (Figure 3C)..

AURKA and KIF2C are thus preferred as they are exclusive for aggressive tumours, providing evidence that they are directly involved in the malignant phenotype (Table 2).

Table 2: Occurrence of overexpression of the candidate biomarkers over the PAM50 breast cancer subtypes expressed as a percentage of the subtype population (N = 525).

PAM50 annotation retrieved from data generated by TCGA Research Network: http://cancergenome.nih.gov/ and data generated from the cBioPortal accessible through http://www.cBioPortal.org/ (Cerami, etal, 2012; Gao, etal, 2013).

PP2A deregulation biomarkers	*Basal (N=99) %*	*HER2-enriched (N=58) %*	*Luminal B (N=133) %*	*Luminal A (N=235) %*
AURKA: EXP>2	47	36	30	3
KIF2C: EXP>2	44	7	5	0
Total cases with deregulation	56	36	30	3

5

KIF2C and AURKA expression also correlated with percentage cell viability (Table 3) during FTY720 treatment in FTY720-sensitive cell lines. There was no correlation, when non sensitive cell lines were used. AURKA and KIF2C also correlates with CIP2A across treatment.. This supports the use of KIF2C and AURKA as biomarkers to identify
10 novel therapeutic groups within breast cancer patients eligible for PP2A activation therapy.

Table 3: Spearman correlation analysis between the expression of AURKA, KIF2C and CIP2A following treatment at 0.1. 1 and 5μM FTY720 in the FTY720-sensitive cell lines. Correlation of expression with cellular viability is included.

The normalised Quantigene expression levels were used.

[N=16 conditions (4 sensitive cell lines at 4 treatment conditions); Sig. (2-tailed) stands for two-tailed significance, significance at the 95% confidence interval is highlighted]

	CIP2A	**KIF2C**	**AURKA**	**Viability (24hours)**

CIP2A	Spearman Correlation	n/a	0.677**	0.808**	0.671**
	Sig. (2-tailed)		0.04	0.0002	0.004
KIF2C	Spearman Correlation	0.677**	n/a	0.887**	0.879**
	Sig. (2-tailed)	0.004		0.000005	0.00001
AURKA	Spearman Correlation	0.808**	0.887**	n/a	0.799**
	Sig. (2-tailed)	0.0002	0.000005		0.0002

Example 3 - PP2A activity biomarkers are correlated with protein expression of inhibitors and pS6K

5

AURKA expression was also found to correlate strongly with cytoplasmic cip2a staining **(Table 4).** AURKA significantly correlates with nuclear pS6K, establishing an association between AURKA expression and growth factor (PBK/mTOR) signaling attenuation as part of the negative feedback driven by PP2A.

10

Table 4: Spearman correlations between PP2A activity biomarker expression and the protein expression of inhibitors ofPP2A and downstream phospho-proteins with defined localisation in the breast cancer cell lines.

*Expression ofPP2A activity biomarkers is normalised to the housekeeping genes, while protein expression was scored using the H-score (0-300)for each cellular compartment and then expressed as positive or negative based on thresholds set on significance with survival and/or correlation with histopathological factors [*significant at 95% confidence interval; + significant at 90% confidence interval]*

Protein / **Biomarker**		**pAKT**		**pS6K**	
		Cytoplasmic	**Nuclear**	**Cytoplasmic**	**Nuclear**
AURKA	Spearman's Rho	0.124	-0.058	0.032	0.272
	P	0.344	0.661	0.806	0.035*
KIF2C	Spearman's Rho	-0.138	-0.238	0.079	0.198
	P	0.293	0.067+	0.548	0.129
Protein / **Biomarker**		Cytoplasmic CIP2A	Nuclear SET	Cytoplasmic SET	Cytoplasmic α4

AURKA	Spearman's Rho	0.378	0.197	0.249	0.295
	P	0.003*	0.132	0.055+	0.025
KIF2C	Spearman's Rho	0.227	0.005	-0.033	0.129
	P	0.083+	0.967	0.800	0.336

Hence, in this study we identified a novel subgroup of basal-type patients, using KIF2C and AURKA as biomarkers of sensitivity to the PP2A activator FTY720. The sensitivity of this class to FTY720 is supported following proper and expected classification of the sensitive cell lines within this subgroup.

FURTHER ASPECTS OF THE INVENTION

1. A method of treating a patient having a cancer comprising an overexpression of AURKA and/or KIF2C, comprising administering to the patient a PP2A activator and thereby treating the cancer.

2. The method according to item 1, wherein said patient has an overexpression of AURKA and KIF2C.

3. A method of treating cancer in a patient, the method comprising administering an AURKA antagonist and/or a KIF2C antagonist to the patient and thereby treating the cancer.

4. The method according to item 3, further comprising administering a PP2A activator.

5. The method according to any one of the preceding items, which is for treating breast cancer.

6. The method according to item 5, which is for treating basal breast cancer.

7. The method according to any one of the preceding items, wherein the cancer has an underexpression of one or more of HER2, ER and PR.

8. A method according to item 7, wherein the cancer has an underexpression of
5 HER2, ER and PR.

9. A method according to any one of the preceding items, wherein the patient is selected for said treatment on the basis of having a cancer comprising an overexpression of AURKA and/or KIF2C.

10

10. A method according to any one of the preceding items, wherein the patient is selected for said treatment on the basis of the cancer having an underexpression of one or more of HER2, ER and PR.

15 11. A method according to item 9 or 10 wherein said selection for treatment is carried out on diagnosis of the cancer, during treatment of the cancer, or following resistance to a cancer therapy.

12. A method according to any one of the preceding items, wherein the PP2A activator
20 is a small molecule, a protein, an antibody, a polynucleotide, an oligonucleotide, an anti sense RNA, a small interfering RNA (siRNA) or a small hairpin RNA (shRNA).

13. A method according to any one of the preceding items, wherein the PP2A activator and/or the AURKA antagonist and/or KIF2C antagonist is administered in combination
25 with another cancer therapy.

14. A method according to any one of the preceding items, wherein the patient is human.

30 15. A method of treating cancer in a patient, the method comprising (a) measuring the amount of AURKA and/or KIF2C in the cancer and (b) if the cancer comprises an

overexpression of AURKA and/or KIF2C, administering to the patient a PP2A activator and thereby treating the cancer.

16. A PP2A activator for use in treating cancer in a patient, wherein the cancer
5 comprises an overexpression of AURKA and/or KIF2C.

17. An AURKA antagonist and/or a KIF2C antagonist for use in treating cancer in a patient.

10 18. Use of a PP2A activator in the manufacture of a medicament for treating cancer in a patient, wherein the cancer comprises an overexpression of AURKA and/or KIF2C.

19. Use of an AURKA antagonist and/or a KIF2C antagonist in the manufacture of a medicament for treating cancer in a patient.

15

20. A method for detecting cancer in a patient, the method comprising measuring expression of AURKA and KIF2C in the patient, wherein overexpression of AURKA and KIF2C indicates that the patient comprises a cancer.

20 21. A method for prognosing a cancer in a patient, the method comprising determining whether or not the cancer comprises an overexpression of AURKA and/or KIF2C, wherein an overexpression of AURKA and/or KIF2C in the cancer indicates that the patient has a worse prognosis than in the situation of normal expression of AURKA and/or KIF2C.

25 22. A method for determining whether or not a patient having or suspected of having or being at risk of developing cancer will respond to treatment with a PP2A activator, which method comprises measuring expression of AURKA and/or KIF2C in the individual, and thereby predicting whether or not the patient will respond to treatment with a PP2A activator.

30

23. A method for classifying a cancer in a patient, the method comprising measuring expression of AURKA and/or KIF2C in the patient, and classifying the cancer as of a particular subtype based on the expression.

5 24. The method according to any one of items 20 to 23, which further comprises measuring expression of one or more of HER2, ER and PR in the patient.

25. The method according to any one of items 20 to 24, which comprises determining whether there is a deregulation of PP2A in the patient.

10

26. The method according to any one of items 20 to 25, wherein the cancer is breast cancer.

27. The method according to item 23 or any of items 24 to 26 as dependent on item 23,
15 comprising classifying the cancer as a luminal or basal breast cancer, wherein overexpression of AURKA and/or KIF2C classifies the cancer as a basal breast cancer, and wherein normal expression of AURKA and/or KIF2C classifies the cancer as a luminal breast cancer.

20 28. The method according to item 27, further comprising measuring expression of one or more of HER2, ER and PR to further classify the cancer.

29. A kit for treating cancer comprising (a) one or more reagents suitable for measuring expression of AURKA and/or KIF2C and (b) a PP2A activator.

25

30. A kit for detecting a cancer comprising a deregulation of PP2A, comprising reagents suitable for detecting expression of AURKA and/or KIF2C.

31. The kit according to item 30, which comprises reagents suitable for detecting
30 expression of AURKA and KIF2C.

32 The kit according to item 30 or 31 further comprising reagents suitable for detecting expression of an endogenous inhibitor of PP2A or a PP2A subunit.

33. The kit according to any one of items 30-32 wherein the reagents suitable for detecting expression are selected from nucleic acid probes or primers and antibodies.

34. A system for detecting, classifying or prognosing cancer in a patient, or for predicting responsiveness of a cancer patient to treatment with a PP2A activator, the system comprising:

(a) a measuring module for determining expression of AURKA and/or KIF2C in the patient,

(b) a storage module configured to store control data and output data from the measuring module,

(c) a computation module configured to provide a comparison between the value of the output data from the measuring module and the control data; and

(d) an output module configured to display whether or not the patient has cancer based on the comparison,

wherein an overexpression of AURKA and/or KIF2C in the patient indicates the presence of cancer, classifies the cancer, indicates a worse prognosis of cancer, or predicts that the patient will respond to treatment with a PP2A activator.

CLAIMS

1, A method of treating a patient having a cancer comprising an overexpression of AURKA and/or KIF2C, comprising administering to the patient a PP2A activator and thereby treating the cancer.

2. The method according to claim 1, wherein said patient has an overexpression of AURKA and KIF2C.

3. The method according to claim 1 or 2, which is for treating breast cancer.

4. The method according to claim 3, which is for treating basal breast cancer.

5. The method according to any one of the preceding claims, wherein the cancer has an underexpression of one or more of HER2, ER and PR.

6. A method according to claim 5, wherein the cancer has an underexpression of HER2, ER and PR.

7. A method according to any one of the preceding claims, wherein the patient is selected for said treatment on the basis of having a cancer comprising an overexpression of AURKA and/or KIF2C.

8. A method according to any one of the preceding claims, wherein the patient is selected for said treatment on the basis of the cancer having an underexpression of one or more of HER2, ER and PR.

9. A method according to claim 7 or 8 wherein said selection for treatment is carried out on diagnosis of the cancer, during treatment of the cancer, or following resistance to a cancer therapy.

10. A method according to any one of the preceding claims, wherein the PP2A activator is a small molecule, a protein, an antibody, a polynucleotide, an oligonucleotide, an antisense RNA, a small interfering RNA (siRNA) or a small hairpin RNA (shRNA).

5 11. A method according to any one of the preceding claims, wherein the PP2A activator is administered in combination with another cancer therapy, optionally with an AURKA antagonist and/or a KIF2C antagonist.

12. A method according to any one of the preceding claims, wherein the patient is
10 human.

13. A method of treating cancer in a patient, the method comprising (a) measuring the amount of AURKA and/or KIF2C in the cancer and (b) if the cancer comprises an overexpression of AURKA and/or KIF2C, administering to the patient a PP2A activator
15 and thereby treating the cancer.

14. A PP2A activator for use in treating cancer in a patient, wherein the cancer comprises an overexpression of AURKA and/or KIF2C.

20 15. Use of a PP2A activator in the manufacture of a medicament for treating cancer in a patient, wherein the cancer comprises an overexpression of AURKA and/or KIF2C.

16. A method for determining whether or not a patient having or suspected of having or being at risk of developing cancer will respond to treatment with a PP2A activator, which
25 method comprises measuring expression of AURKA and/or KIF2C in the individual, and thereby predicting whether or not the patient will respond to treatment with a PP2A activator.

17. A method for classifying a cancer in a patient, the method comprising measuring
30 expression of AURKA and/or KIF2C in the patient, and classifying the cancer as of a particular subtype based on the expression.

18. The method according to claim 16 or 17, which further comprises measuring expression of one or more of HER2, ER and PR in the patient.

5 19. The method according to any one of claims 16 to 18, which comprises determining whether there is a deregulation of PP2A in the patient.

20. The method according to any one of claims 16 to 19, wherein the cancer is breast cancer.

10

21. The method according to claim 17 or any of claims 18 to 20 as dependent on claim 17, comprising classifying the cancer as a luminal or basal breast cancer, wherein overexpression of AURKA and/or KIF2C classifies the cancer as a basal breast cancer, and wherein normal expression of AURKA and/or KIF2C classifies the cancer as a luminal
15 breast cancer.

22. The method according to claim 21, further comprising measuring expression of one or more of HER2, ER and PR to further classify the cancer.

20 23. A kit for treating cancer comprising (a) one or more reagents suitable for measuring expression of AURKA and/or KIF2C and (b) a PP2A activator.

24. A kit for detecting a cancer comprising a deregulation of PP2A, comprising reagents suitable for detecting expression of AURKA and/or KIF2C.

25

25. The kit according to claim 24, which comprises reagents suitable for detecting expression of AURKA and KIF2C.

26 The kit according to claim 24 or 25 further comprising reagents suitable for
30 detecting expression of an endogenous inhibitor of PP2A or a PP2A subunit.

27. The kit according to any one of claims 24-26 wherein the reagents suitable for detecting expression are selected from nucleic acid probes or primers and antibodies.

28. A system for classifying cancer in a patient, or for predicting responsiveness of a
5 cancer patient to treatment with a PP2A activator, the system comprising:

(a) a measuring module for determining expression of AURKA and/or KIF2C in the patient,

(b) a storage module configured to store control data and output data from the measuring module,

10 (c) a computation module configured to provide a comparison between the value of the output data from the measuring module and the control data; and

(d) an output module configured to display whether or not the patient has cancer based on the comparison,

wherein an overexpression of AURKA and/or KIF2C in the patient classifies the
15 cancer, or predicts that the patient will respond to treatment with a PP2A activator.

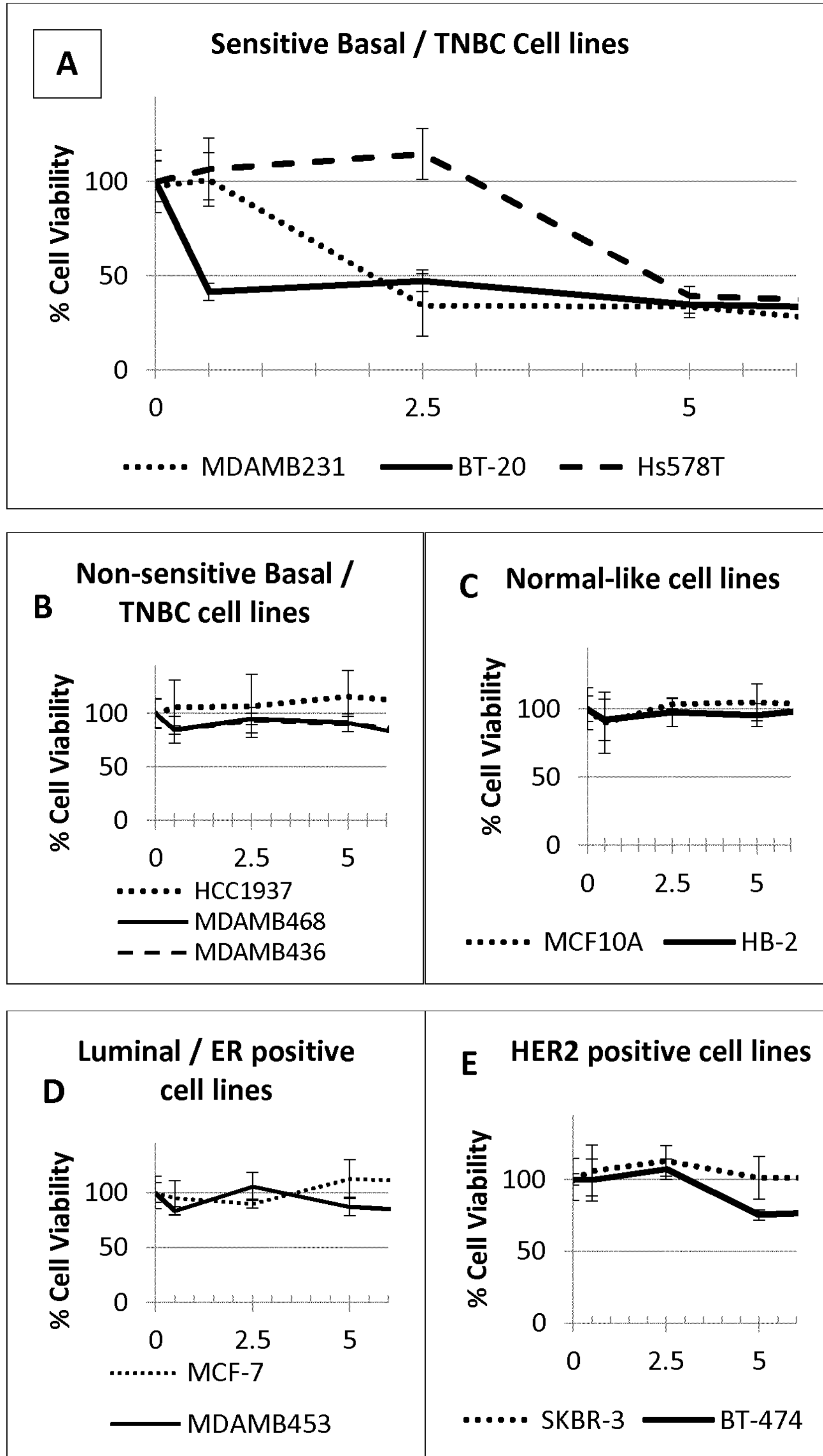
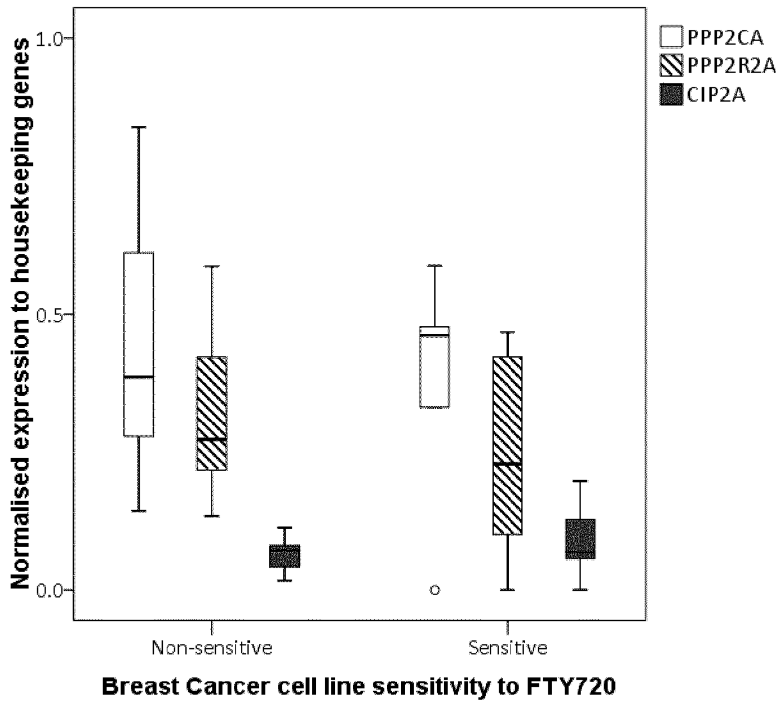


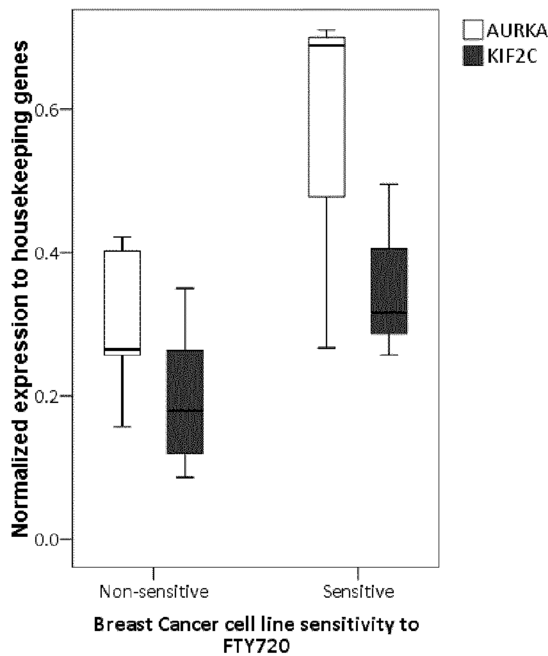
Figure 1



Distribution of PP2A complex and regulator expression between sensitive and non-sensitive cell lines

Gene	Mann-Whitney U test, *P*
PPP2CA	0.256
PPP2R2A	0.310
CIP2A	0.040*

Figure 2A.



Distribution of PP2A complex and regulator expression between sensitive and non-sensitive cell lines

Candidate Biomarkers	Mann-Whitney U test, *P*
AURKA	0.014*
KIF2C	< 0.001*

Figure 2B

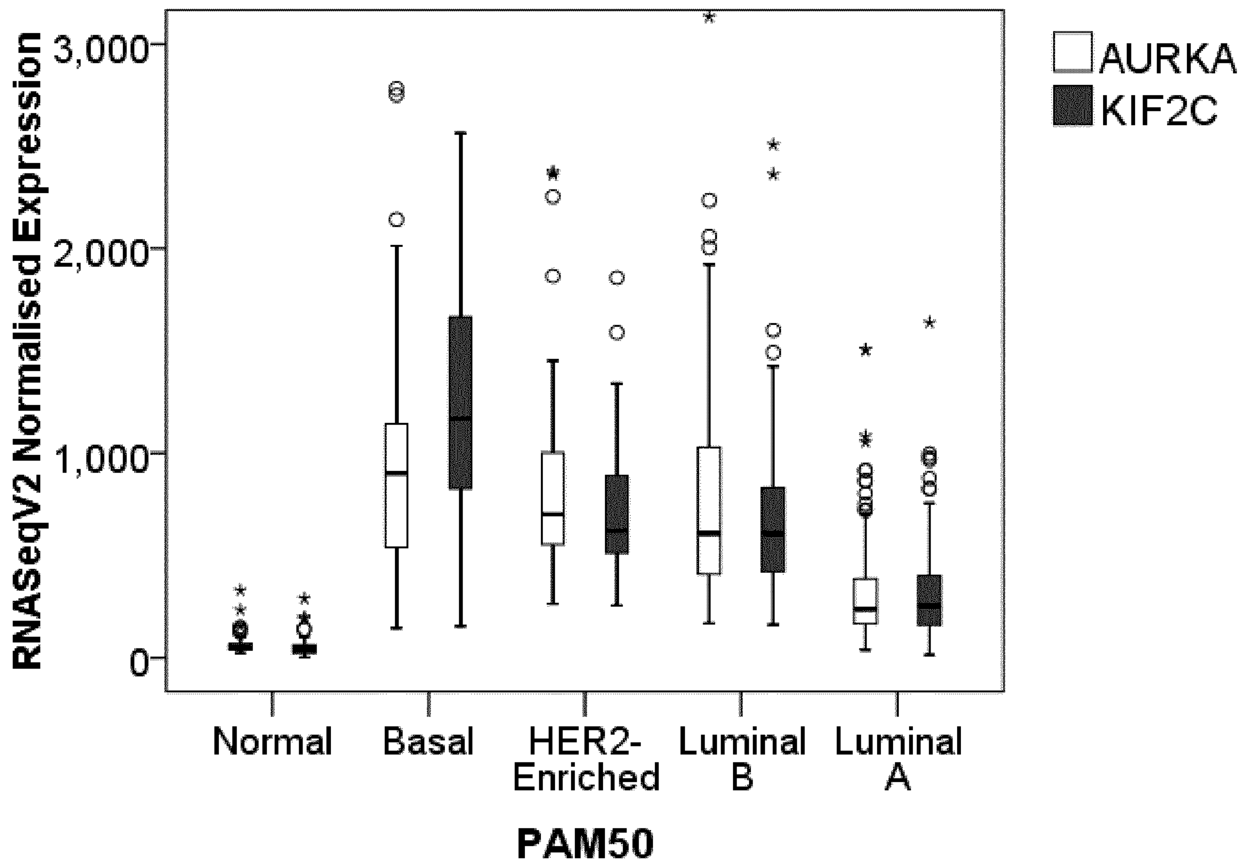
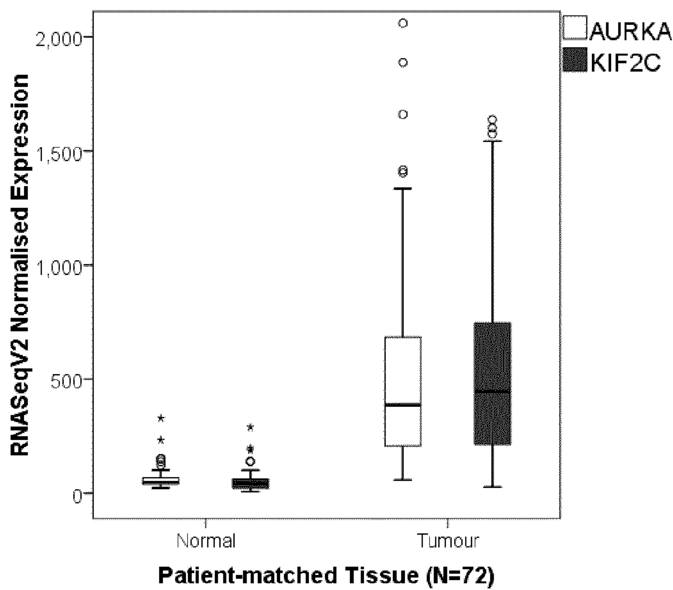


Figure 3A



Candidate Biomarkers	*Wilcoxon Signed Rank test, P*
AURKA	< 0.001*
KIF2C	< 0.001*

Figure 3B

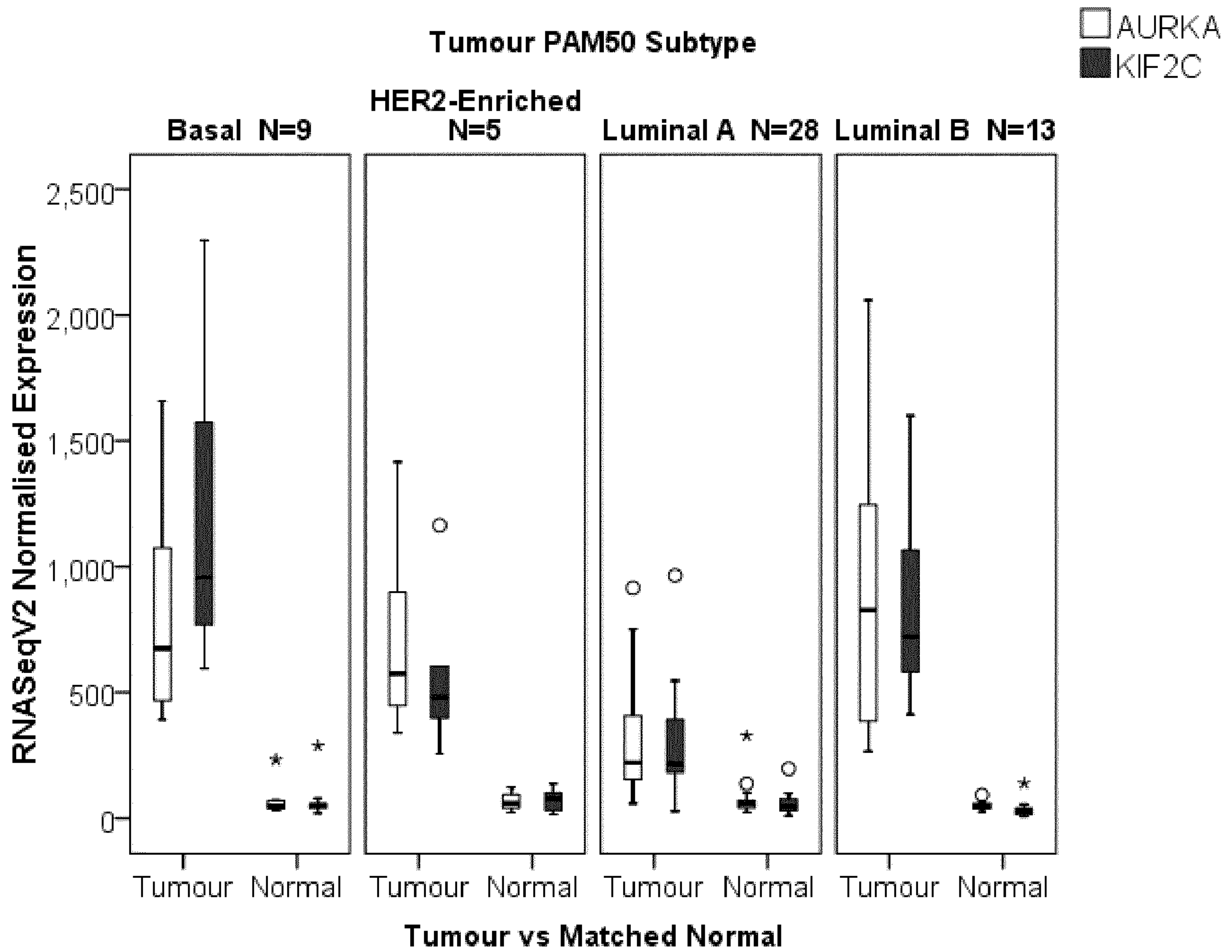


Figure 3C

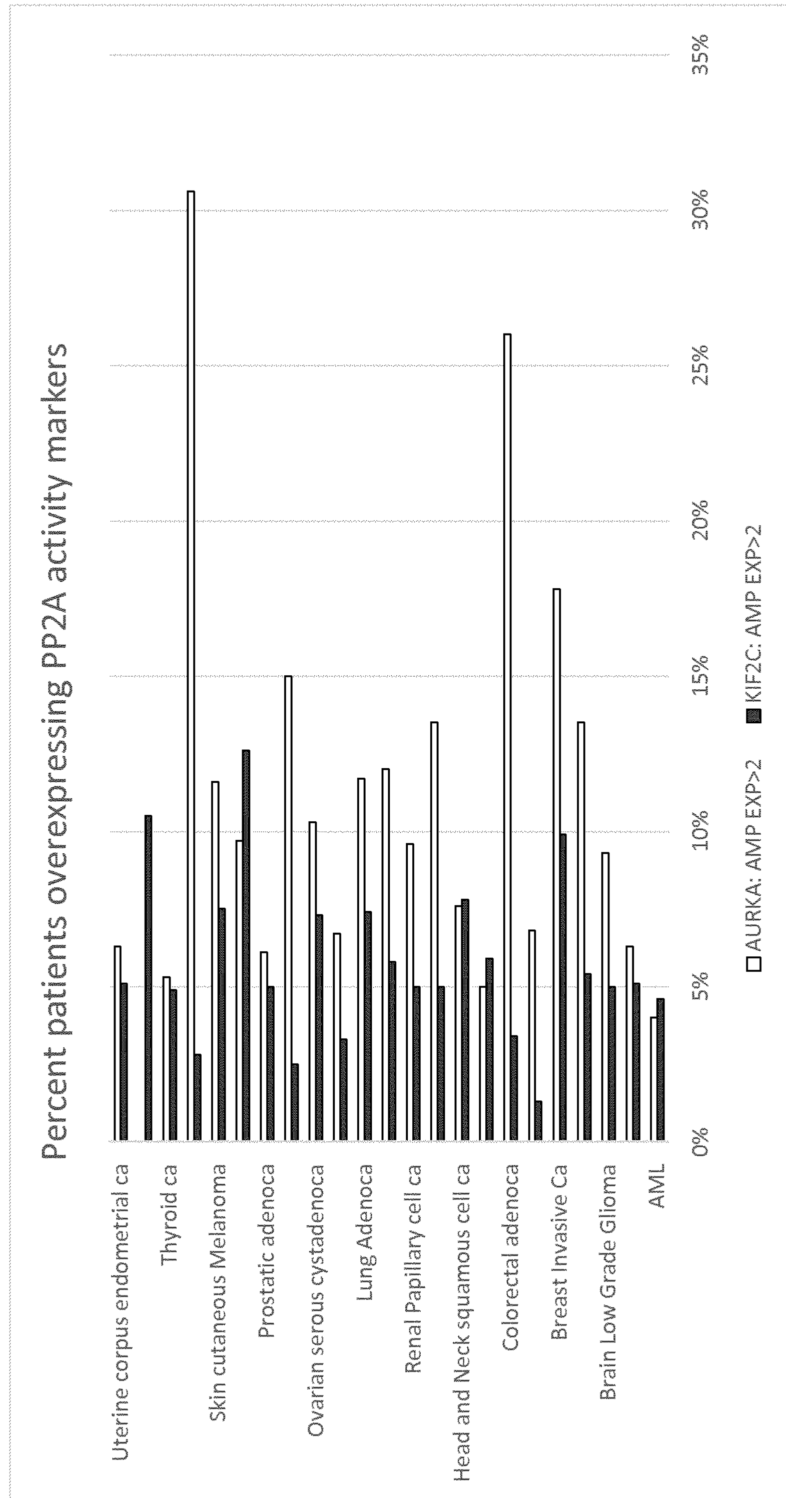


Figure 4.

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2018/057189

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61P35/00 A61K31/137 G01N33/50 A61K38/00 A61K39/00
ADD.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
A61P A61K G01N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

EPO-Internal , WPI Data, BIOSIS, EMBASE, CHEM ABS Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2011/153545 A2 (BIOARRAY THERAPEUTICS INC [US]; MARTIN KATHERINE J [US]; MAGEE DAVID [ ]) 8 December 2011 (2011-12-08)	1,3, 6-18, 20-24,27
Y	paragraphs [0161], [0171]; claims	1-28
X	WO 2010/096574 A1 (LISANTI MICHAEL P [US]; SOTGIA FEDERICA [US]; PESTELL RICHARD G [US]) 26 August 2010 (2010-08-26)	1,3,5, 7-10, 12-16, 18,20, 23,24,27
Y	claims	1-28
X	WO 2008/077165 A1 (ARC AUSTRIAN RES CENTERS GMBH [AT]; LAUSS MARTIN [AT]; VIERLINGER KLEM) 3 July 2008 (2008-07-03) claims	17,20, 23-25,27

-/-

[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance
"E" earlier application or patent but published on or after the international filing date
"L" documentwhich may throw doubts on priority claim(s) orwhich is cited to establish the publication date of another citation or other special reason (as specified)
"O" document referring to an oral disclosure, use, exhibition or other means
"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
18 June 2018	06/07/2018

Name and mailing address of the ISA/
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Langer, Astrid

1

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	KATSHA AHMED M ET AL: "Aurora Kinase A Promotes Resistance to Everolimus Through Regulation of PP2A-eIF4E Axis in Upper Gastrointestinal Cancers", GASTROENTEROLOGY, vol. 150, no. 4, Suppl. 1, April 2016 (2016-04), page S187, XP055485201, & DIGESTIVE DISEASE WEEK (DDW); SAN DIEGO, CA, USA; MAY 20 -24, 2016 abstract -----	1-28
Y	CAI FEN ET AL: "Cucurbitacin B reverses multidrug resistance by targeting CIP2A to reactivate protein phosphatase 2A in MCF-7/Adriamycin cells", ONCOLOGY REPORTS, vol. 36, no. 2, August 2016 (2016-08), pages 1180-1186, XP55485007, abstract; figures -----	1-28
Y	TSENG LING-MING ET AL: "Erlotinib derivative, devoid of EGFR kinase inhibiting effect, induced apoptosis of triple negative breast cancer cells through modulating Elk-l/CIP2A signaling pathway", CANCER RESEARCH, vol. 76, no. Suppl. 14, July 2016 (2016-07), page 3065, XP55485009, & 107TH ANNUAL MEETING OF THE AMERICAN-ASSOCIATION-F0R-CANCER-RESEARCH (AACR); NEW ORLEANS, LA, USA; APRIL 16 -20, 2016 abstract -----	1-20
Y	RINCON RAUL ET AL: "PP2A inhibition determines poor outcome and doxorubicin resistance in early breast cancer and its activation shows promising therapeutic effects", 0NC0TARGET, vol. 6, no. 6, February 2015 (2015-02), pages 4299-4314, XP55480107, abstract; figures -----	1-20

-/--

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
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