VHZ FOR DIAGNOSIS AND TREATMENT OF CANCER

FIELD

The present invention relates to the fields of medicine, cell biology, molecular biology and genetics. This invention relates to the field of medicine. In particular, it relates to treatment and diagnosis of diseases such as breast cancer, as well as compositions for such use.

BACKGROUND

VHZ is a phosphatase that shares about 28% amino acid sequence identity with human PRL-PTPs. VHZ was previously reported to be expressed in many tissues and located in the cytosol and in nucleoli (Alonso et al., 2004a).

However, the role of VHZ was largely unknown; despite its conservation through evolution with orthologues in frogs, fish, fly, and the Archaea. VHZ, as well as VHR, belongs to a separate subgroup of VH1-like PTPs (Alonso et al, 2004b). VHR has been reported to have a function in regulating cell cycle progression (Rahmouni et al., 2006).

In the Western world and the developed countries of Asia, breast carcinoma is the second leading cause of cancer-related death in women (Polyak, 2001). Breast cancer tops the cancer list for women in Singapore, with 700-800 new cases being diagnosed each year (Singapore Cancer Registry Report, 2000). In the USA, 180,000 women are diagnosed annually with new cases of breast cancer (Polyak, 2001). Despite better diagnosis and routine screening around a quarter of the cases will die from their disease.

Accordingly, there is a need for improved breast cancer detection and therapy.

SUMMARY

In one aspect, the present invention refers to the use of an anti-VHZ agent in the manufacture of a medicament for the treatment, prophylaxis or alleviation of breast cancer, wherein the breast cancer is not an invasive or metastatic breast cancer, and wherein VHZ is overexpressed in the breast cancer.

In one example, there is disclosed ~~According to a 1st aspect of the present invention, we provide~~ VHZ for use in a method of treatment, prophylaxis or alleviation of a cancer, such as breast cancer, in an individual.

In another example, there is described ~~There is provided, according to a 2nd aspect of the present invention,~~ an anti-VHZ agent for the treatment, prophylaxis or alleviation of cancer. The cancer may comprise breast cancer. The cancer may comprise an invasive or metastatic cancer such as Invasive Ductal Carcinoma (IDC).

The anti-VHZ agent may be capable of down-regulating any combination of the expression, amount or activity of a VHZ sequence shown as GenBank accession number NM_017823 or NP_060293, or a sequence which has at least 90% sequence identity to that sequence. The anti-VHZ agent may comprise an anti-VHZ antibody.

The anti-VHZ antibody may comprise an anti-peptide antibody generated against RRLRPGSIETYEQEK corresponding to amino acid residues (126-140) of human VHZ.

The anti-VHZ antibody may comprise chicken anti-human VHZ antibody (catalogue numbers LS-C32281, amino acids 35 to 90, LS-C42458, LS-A6806 and LS-A6803, LS-C32281, LifeSpan Inc, Seattle, Washington, USA), rabbit anti-human VHZ antibody (catalogue number DS-PB-00676, RayBiotech Inc, Norcross, Georgia, USA), chicken anti-human VHZ antibody (catalogue number XW-7857, ProSci Incorporated, Poway, California, USA), rabbit anti-human VHZ antibody (catalogue number F4560 and D9840-66A, United States Biological, Swampscott, Massachusetts, USA), chicken anti-human VHZ antibody (catalogue number D9840-66, United States Biological, Swampscott, Massachusetts, USA), rabbit anti-human VHZ antibody (catalogue number AHP1142, AdB Serotec, Oxford, United Kingdom), rabbit anti-human VHZ antibody (catalogue number NB110-40452, Novus Biologicals, Littleton, Colorado, USA), chicken anti-human VHZ antibody (catalogue number NB100-75328, Novus Biologicals, Littleton, Colorado, USA).

The anti-VHZ agent may be capable of downregulating VHZ by RNA interference. It may comprise a Small Interfering RNA (siRNA), Short Hairpin RNA (shRNA) or Chimera RNAi such as a DUSP23 Pre-design Chimera RNAi (catalogue number H00054935-R01, Novus Biologicals, Littleton, Colorado, USA).

In yet another example, there is disclosed~~We provide, according to a 3rd aspect of the present invention,~~ a kit for detecting breast cancer in an individual or susceptibility of the individual to breast cancer. The kit may comprise means for detection of VHZ expression in the individual or a sample taken from him or her. The means for detection may be selected from the group consisting of: a VHZ polynucleotide or a fragment thereof; a complementary nucleotide sequence to VHZ nucleic acid or a fragment thereof; a VHZ polypeptide or a fragment thereof, or an anti-VHZ antibody, or an anti-VHZ agent as set out above, and optionally instructions for use. It may further comprise a therapeutic drug for treatment, prophylaxis or alleviation of breast cancer, such as comprising Tamoxifen or Herceptin.

In a further example, there is described~~As a 4th aspect of the present invention, there is provided~~ a method of detecting a cancer cell such as a breast cancer cell. The cancer cell may comprise invasive or metastatic cancer cell such as Invasive Ductal Carcinoma (IDC). The method may comprise detecting modulation of expression, amount or activity of VHZ in the cell. The modulation may comprise up-regulation. The expression of VHZ may be compared to the expression, amount or activity of VHZ in a control cell known to be non-cancerous.

The method may comprise detecting a VHZ nucleic acid. This may be by means of a probe comprising at least a portion of a nucleic acid having a sequence shown as GenBank accession number NM_017823 or NP_060293 or a sequence having at least 90% sequence identity to such a sequence, or in which the method comprises detecting a VHZ polypeptide, such as by means of an anti-VHZ antibody set out in Claim 2.

The method may further comprise histological grading. The histological grading may be by means of the Elston-Ellis modified Scarff, Bloom, Richardson grading system (Nottingham Grading System (NGS)).

In another example, there is described~~We provide, according to a 5th aspect of the present invention,~~ a method of determining the proliferative state of a cell, or determining the likelihood that a cell will become invasive or aggressive. The method comprises detecting modulation of expression, amount or activity of VHZ in the cell.

In yet another example, there is described~~The present invention, in a 6th aspect, provides~~ a method of predicting a survival rate of an individual with cancer. The method comprises detecting modulation of expression of VHZ in a cell of the individual

In yet another example, there is described~~In a 7th aspect of the present invention, there is provided~~ a method of choosing a therapy for an individual with cancer, the method comprising detecting modulation of expression of VHZ in a cell of the individual choosing an appropriate therapy based on the aggressiveness of the cancer. The therapy may comprise an anti-VHZ agent as described above.

In a further example, there is described~~According to an 8th aspect of the present invention, we provide~~ a method of determining the likelihood of success of a particular therapy in an individual with a cancer. The method comprises comparing the therapy with a therapy determined by a method as set out above.

Each of these methods may further comprise a feature set out above in any of the examples~~1st to 3rd aspects of the invention~~.

In yet another example, there is described~~We provide, according to a 9th aspect of the invention,~~ a method of manipulating a cancer cell, such as a breast cancer cell. The cancer cell may comprise an invasive or metastatic cancer cell such as Invasive Ductal Carcinoma (IDC). The method may comprise modulating the expression, amount or activity of VHZ in the cell. The modulation may comprise down-regulation. The method may comprise exposing the cell to an siRNA or shRNA capable of specifically binding to VHZ. It may comprise exposing the cell to an anti-VHZ antibody such as set out above. The cancer cell may become non-cancerous or the invasive or metastatic cancer cell may become non-invasive or non-metastatic as a result of the manipulation.

In one example, there is described~~There is provided, in accordance with a 10th aspect of the present invention,~~ a method of manipulating a cell, the method comprising the steps of: (a) detecting increased VHZ expression, amount or activity in a cell; and (b) reducing the level of VHZ in the cell.

In another example, there is described~~As an 11th aspect of the invention, we provide~~ a method of identifying a molecule capable of binding to a VHZ polypeptide, the method comprising contacting a VHZ polypeptide or a sequence having at least 90% sequence identity thereto with a candidate molecule and determining whether the candidate molecule binds to the VHZ polypeptide or sequence having at least 90% sequence identity thereto.

In yet another example, there is described~~We provide, according to a 12th aspect of the invention, there is provided~~ a method of identifying a modulator of VHZ, the method comprising contacting a cell with a candidate molecule and detecting elevated or reduced expression, amount or activity of VHZ in or of the cell.

In a further example, there is described~~According to a 13th aspect of the present invention, we provide~~ a method of identifying a molecule suitable for the treatment, prophylaxis or alleviation of cancer, the method comprising determining if a candidate molecule is an agonist or antagonist of VHZ or a sequence having at least 90% sequence identity thereto. The method may comprise exposing a candidate molecule to a VHZ polypeptide or a cell expressing a VHZ polypeptide in order to determine if the candidate molecule is an agonist or antagonist thereof.

In yet another example, there is described~~There is provided, according to a 14th aspect of the present invention,~~ a method of identifying an agonist or antagonist of a VHZ or a sequence having at least 90% sequence identity thereto, the method comprising administering a candidate molecule to an animal and determining whether the animal exhibits increased or decreased expression, amount or activity of VHZ.

In one example, there is described~~We provide, according to a 15th aspect of the present invention,~~ a method of treatment, prophylaxis or alleviation of a cancer in an individual, the method comprising modulating the expression, amount or activity of a VHZ in a cell of an individual. The cancer may comprise breast cancer, such as invasive or metastatic cancer such as Invasive Ductal Carcinoma (IDC). The method may be such that the expression, amount or activity of VHZ is decreased in a breast cell of the individual.

In one example, there is described~~We provide, according to a 16th aspect of the present invention,~~ a method of diagnosis of a cancer or susceptibility to cancer in an individual or prognosis of an individual with cancer, the method comprising detecting modulation of expression, amount or activity of VHZ in a cell of the individual. The cancer may comprise breast cancer, such as invasive or metastatic cancer such as Invasive Ductal Carcinoma (IDC).

In another example, there is described~~We provide, according to a 17th aspect of the present invention,~~ a method of determining whether a tumour in an individual is, or is likely to be, an invasive or metastatic tumour, the method comprising detecting modulation of expression, amount or activity of VHZ in a tumour cell of the individual.

In yet another example, there is described~~We provide, according to a 18th aspect of the present invention,~~ a method of treatment, prophylaxis or alleviation of cancerin an individual, the method comprising detecting modulation of expression, amount or activity of VHZ in a cell of the individual and administering an appropriate therapy to the individual based on the aggressiveness of the tumour. The therapy may comprise an anti-VHZ agent as described above. The cancer may comprise breast cancer, such as invasive or metastatic cancer such as Invasive Ductal Carcinoma (IDC).

The diagnosis, prognosis or choice of therapy may be further determined by assessing the size of the tumour, or the lymph node stage, or both, optionally together or in combination with other risk factors. The diagnosis, prognosis or choice of therapy may be further determined by assessing the oestrogen receptor (ER) status of the tumour.

In another example, there is disclosed~~We provide, according to a 19th aspect of the present invention,~~ molecule, agonist or antagonist of a VHZ polypeptide identified by a method or use as set out above.

In another example, there is disclosed~~We provide, according to a 20th aspect of the present invention,~~ a molecule capable of modulating, such as down-regulating, the expression of a VHZ for use in the treatment, prophylaxis or alleviation of cancer. The molecule may comprise an anti-peptide antibody generated against RRLRPGSIETYEQEK corresponding to amino acid residues (126-140) of human VHZ.

The practice of the present invention will employ, unless otherwise indicated, conventional techniques of chemistry, molecular biology, microbiology, recombinant DNA and immunology, which are within the capabilities of a person of ordinary skill in the art. Such techniques are explained in the literature. See, for example, J. Sambrook, E. F. Fritsch, and T. Maniatis, 1989, *Molecular Cloning: A Laboratory Manual*, Second Edition, Books 1-3, Cold Spring Harbor Laboratory Press; Ausubel, F. M. et al. (1995 and periodic supplements; *Current Protocols in Molecular Biology*, ch. 9, 13, and 16, John Wiley & Sons, New York, N.Y.); B. Roe, J. Crabtree, and A. Kahn, 1996, *DNA Isolation and Sequencing: Essential Techniques*, John Wiley & Sons; J. M. Polak and James O'D. McGee, 1990, *In Situ Hybridization: Principles and Practice*; Oxford University Press; M. J. Gait (Editor), 1984, *Oligonucleotide Synthesis: A Practical Approach*, Irl Press; D. M. J. Lilley and J. E. Dahlberg, 1992, *Methods of Enzymology: DNA Structure Part A: Synthesis and Physical Analysis of DNA* Methods in Enzymology, Academic Press; Using Antibodies : A Laboratory Manual : Portable Protocol NO. I by Edward Harlow, David Lane, Ed Harlow (1999, Cold Spring Harbor Laboratory Press, ISBN 0-87969-544-7); Antibodies : A Laboratory Manual by Ed Harlow (Editor), David Lane (Editor) (1988, Cold Spring Harbor Laboratory Press, ISBN 0-87969-314-2), 1855. Handbook of Drug Screening, edited by Ramakrishna Seethala, Prabhavathi B. Fernandes (2001, New York, NY, Marcel Dekker, ISBN 0-8247-0562-9); and Lab Ref: A Handbook of Recipes, Reagents, and Other Reference Tools for Use at the Bench, Edited Jane Roskams and Linda Rodgers, 2002, Cold Spring Harbor Laboratory, ISBN 0-87969-630-3. Each of these general texts is herein incorporated by reference.

SEQUENCE LISTING

<110> Agency For Science, Technology and Research (A*STAR)
Zeng, Qi

<120> VHZ for Diagnosis and Treatment of Cancer

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