

R-SPONDIN TRANSLOCATIONS AND METHODS USING THE SAME

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application claims benefit under 35 U.S.C. § 119 to United States Patent Application No. 61/597,746 filed on February 11, 2012 and 61/674,763 filed on July 23, 2012, the entire contents of which are incorporated herein by reference.

SEQUENCE LISTING

[0002] The instant application contains a Sequence Listing submitted via EFS-Web and hereby incorporated by reference in its entirety. Said ASCII copy, created on January 31, 2013, is named P4853R1WO_PCTSequenceListing.txt and is 58,980 bytes in size.

FIELD

[0003] Provided are therapies related to the treatment of pathological conditions, such as cancer.

BACKGROUND

[0004] Colorectal cancer (CRC) with over 100,000 new cases reported annually is the fourth most prevalent cancer and accounts for over 50,000 deaths per year in the United States (Siegel, R. *et al.*, *CA: A Cancer Journal for Clinicians* 61:212-236 (2011)). Approximately 15% of CRCs exhibit microsatellite instability (MSI) arising from defects in DNA mismatch repair (MMR) system (Fearon, E. R., *Annu. Rev. Pathol.* 6:479-507 (2011)). The other ~85% of microsatellite stable (MSS) CRCs are the result of chromosomal instability (CIN) (Fearon, E. R., *Annu. Rev. Pathol.* 6:479-507 (2011)). Genomic studies have identified acquisition of mutations in genes like *APC*, *KRAS*, and *TP53* during CRC progression (Fearon, E. R., *Annu. Rev. Pathol.* 6:479-507 (2011)). Sequencing colon cancer protein-coding exons and whole genomes in a small number of samples have identified several additional mutations and chromosomal structural variants that likely contribute to oncogenesis (Wood, L. D. *et al.*, *Science* 318:1108-1113 (2007); Timmermann, B. *et al.*, *PloS One* 5:e15661 (2010)). However, recent insertional mutagenesis screens in mouse models of colon cancer suggested involvement of additional genes and pathways in CRC development (Starr, T. K. *et al.*, *Science* 323:1747-1750 (2009); March, H. N. *et al.*, *Nat. Genet.* 43:1202-1209 (2011)).

[0005] There remains a need to better understand the pathogenesis of cancers, in particular, human colon cancers and also to identify new therapeutic targets.

SUMMARY

[0006] The disclosure provides wnt pathway antagonists including R-spondin-translocation antagonists and methods of using the same.

[0006a] In one aspect, there is provided an *in vitro* method of inhibiting proliferation of a cancer cell comprising contacting the cancer cell with an effective amount of an R-spondin-translocation

antagonist specific for R-spondin chromosomal translocation.

[0006b] In one aspect, there is provided the use of an effective amount of an ~~R-spondin-translocation~~ antagonist specific for R-spondin chromosomal translocation in the manufacture of a medicament for inhibiting proliferation of a cancer cell *in vivo* wherein said cancer cell is to be contacted with said medicament.

[0007] Provided herein are methods of inhibiting cell proliferation of a cancer cell comprising contacting the cancer cell with an effective amount of an R-spondin-translocation antagonist.-

Further provided herein are methods of treating cancer in an individual comprising administering to the individual an effective amount of an R-spondin-translocation antagonist. In some embodiments of any of the methods, the cancer or cancer cell comprises an R-spondin translocation.

[0007a] In one aspect, there is provided the use of an effective amount of an ~~R-spondin~~-antagonist specific for R-spondin in the manufacture of a medicament for treating cancer in an individual identified to have an R-spondin chromosomal translocation, wherein treatment is based upon the individual having cancer comprising an R-spondin chromosomal translocation.

[0007b] In one aspect, there is provided the use of an effective amount of an ~~R-spondin~~-antagonist specific for R-spondin in the manufacture of a medicament for treating a cancer cell identified to have an R-spondin chromosomal translocation in vivo, wherein the cancer cell comprises an R-spondin chromosomal translocation.

[0007c] In one aspect, there is provided the use of an effective amount of an ~~R-spondin~~-antagonist specific for R-spondin in the manufacture of a medicament for treating cancer in an individual provided that the individual has been found to have cancer comprising an R-spondin chromosomal translocation.

[0008] Provided herein are methods of treating cancer in an individual comprising administering to the individual an effective amount of a wnt pathway antagonist, wherein treatment is based upon the individual having cancer comprising an R-spondin translocation. Provided herein are methods of treating a cancer cell, wherein the cancer cell comprises an R-spondin translocation, and wherein the method comprises providing an effective amount of a wnt pathway antagonist. Also provided herein are *methods of treating cancer in an individual provided that the individual has been found to have cancer comprising an R-spondin translocation, the treatment comprising administering to the individual an effective amount of a wnt pathway antagonist.*

[0008a] In one aspect, there is provided the use of an effective amount of an anti-cancer therapy comprising an ~~R-spondin~~-antagonist specific for R-spondin in the manufacture of a medicament for treating cancer in an individual, wherein a sample obtained from the individual has been determined to comprise an R-spondin chromosomal translocation, whereby the cancer is treated.

[0009] Further, provided herein are methods for treating cancer in an individual, the method comprising: determining that a sample obtained from the individual comprises an R-spondin translocation, and administering an effective amount of an anti-cancer therapy comprising a wnt pathway antagonist to the individual, whereby the cancer is treated.

[0009a] In one aspect, there is provided the use of an effective amount of an ~~R-spondin~~-antagonist specific for R-spondin in the manufacture of a medicament for treating cancer, wherein the medicament is to be administered to an individual having cancer that has been identified to have an R-spondin chromosomal translocation and has been selected, wherein the cancer comprises an R-spondin chromosomal translocation, whereby the cancer is treated.

[0010] Provided herein are methods of treating cancer, comprising: (a) selecting an individual having cancer, wherein the cancer comprising an R-spondin translocation; and (b) administering to

the individual thus selected an effective amount of a wnt pathway antagonist, whereby the cancer is treated.

[0011] In one aspect, there is provided a method of identifying an individual with cancer who is more likely or less likely to exhibit benefit from treatment with an anti-cancer therapy comprising an ~~R-spondin~~-antagonist specific for R-spondin, the method comprising: determining presence or absence of an R-spondin chromosomal translocation in a sample obtained from the individual, wherein presence of the R-spondin chromosomal translocation in the sample indicates that the individual is more likely to exhibit benefit from treatment with the anti-cancer therapy comprising the ~~R-spondin~~ antagonist specific for R-spondin or absence of the R-spondin chromosomal translocation indicates that the individual is less likely to exhibit benefit from treatment with the anti-cancer therapy comprising the ~~R-spondin~~-antagonist specific for R-spondin. In some embodiments, the method further comprises administering an effective amount of the anti-cancer therapy comprising a wnt pathway antagonist.

[0012] In one aspect, there is provided a method for predicting whether an individual with cancer is more or less likely to respond effectively to treatment with an anti-cancer therapy comprising an ~~R-spondin~~ antagonist specific for R-spondin, the method comprising determining an R-spondin chromosomal translocation, whereby presence of the R-spondin chromosomal translocation indicates that the individual is more likely to respond effectively to treatment with the ~~R-spondin~~-antagonist specific for R-spondin and absence of the R-spondin chromosomal translocation indicates that the individual is less likely to respond effectively to treatment with the ~~R-spondin~~-antagonist specific for R-spondin. In

some embodiments, the method further comprises administering an effective amount of the anti-cancer therapy comprising a wnt pathway antagonist.

[0013] In one aspect, there is provided a method of predicting the response or lack of response of an individual with cancer to an anti-cancer therapy comprising an ~~R-spondin~~ antagonist specific for R-spondin comprising detecting in a sample obtained from the individual presence or absence of an R-spondin chromosomal translocation, wherein presence of the R-spondin chromosomal translocation is predictive of response of the individual to the anti-cancer therapy comprising the ~~R-spondin~~ antagonist specific for R-spondin and absence of the R-spondin chromosomal translocation is predictive of lack of response of the individual to the anti-cancer therapy comprising the ~~R-spondin~~ antagonist specific for R-spondin. In some embodiments, the method further comprises administering an effective amount of the anti-cancer therapy comprising a wnt pathway antagonist.

[0013a] In one aspect, there is provided the use of an effective amount of an ~~R-spondin-translocation~~ antagonist specific for R-spondin-translocation in the manufacture of a medicament for treating cancer in an individual.

[0014] In some embodiments of any of the methods, the R-spondin translocation is a RSPO1 translocation, RSPO2 translocation, RSPO3 translocation and/or RSPO4 translocation. In some embodiments, the R-spondin translocation is a RSPO2 translocation. In some embodiments, the RSPO2 translocation comprises *EIF3E* and *RSPO2*. In some embodiments, the RSPO2 translocation comprises *EIF3E* exon 1 and *RSPO2* exon 2. In some embodiments, the RSPO2 translocation comprises *EIF3E* exon 1 and *RSPO2* exon 3. In some embodiments, the RSPO2 translocation comprises SEQ ID NO:71. In some embodiments, the R-spondin translocation is a RSPO3 translocation. In some embodiments, the RSPO3 translocation comprises *PTPRK* and *RSPO3*. In some embodiments, the RSPO3 translocation comprises *PTPRK* exon 1 and *RSPO3* exon 2. In some embodiments, the RSPO3 translocation comprises *PTPRK* exon 7 and *RSPO3* exon 2. In some embodiments, the RSPO3 translocation comprises SEQ ID NO:72 and/or SEQ ID NO:73. In some embodiments of any of the methods, the R-spondin translocation is detected at the chromosomal level (e.g., FISH), DNA level, RNA level (e.g., RSPO1-translocation fusion transcript), and/or protein level (e.g., RSPO1-translocation fusion polypeptide).

[0015] In some embodiments of any of the methods, the cancer is colorectal cancer. In some embodiments, the cancer is a colon cancer or rectal cancer.

1) In some embodiments of any of the methods, the wnt pathway antagonist is an antibody, binding polypeptide, small molecule, or polynucleotide. In some embodiments, the wnt pathway antagonist is an R-spondin antagonist. In some embodiments, the R-spondin antagonist is a RSPO1 antagonist, RSPO2 antagonist, RSPO3 antagonist, and/or RSPO4 antagonist. In some embodiments, the wnt pathway antagonist is an isolated monoclonal antibody which binds R-spondin. In some embodiments, the R-spondin is RSPO2 and/or RSPO3. In some embodiments, the R-spondin antagonist is an R-spondin-translocation antagonist. In some embodiments, the R-spondin-translocation antagonist binds a RSPO1-translocation fusion polypeptide and/or polynucleotide, RSPO2-translocation fusion polypeptide and/or polynucleotide, RSPO3-translocation fusion polypeptide and/or polynucleotide

and/or RSPO4-translocation fusion polypeptide and/or polynucleotide. In some embodiments, the R-spondin-translocation antagonist binds a RSPO2-translocation fusion polypeptide and/or polynucleotide. In some embodiments, the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises *EIF3E* and *RSPO2*. In some embodiments, the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises *EIF3E* exon 1 and *RSPO2* exon 2. In some embodiments, the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises *EIF3E* exon 1 and *RSPO2* exon 3. In some embodiments, the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises SEQ ID NO:71. In some embodiments, the R-spondin-translocation fusion polypeptide and/or polynucleotide is a RSPO3-translocation fusion polypeptide and/or polynucleotide. In some embodiments, the RSPO3-translocation fusion polypeptide and/or polynucleotide comprises *PTPRK* and *RSPO3*. In some embodiments, the RSPO3-translocation fusion polypeptide and/or polynucleotide comprises *PTPRK* exon 1 and *RSPO3* exon 2. In some embodiments, the RSPO3-translocation fusion polypeptide and/or polynucleotide comprises *PTPRK* exon 7 and *RSPO3* exon 2. In some embodiments, the RSPO3-translocation fusion polypeptide and/or polynucleotide comprises SEQ ID NO:72 and/or SEQ ID NO:73. In some embodiments, the method further comprises an additional therapeutic agent.

[0016] In one aspect, there is provided an isolated ~~R-spondin-translocation~~ antagonist specific for R-spondin chromosomal translocation, wherein the R-spondin-chromosomal translocation antagonist is an antibody, binding polypeptide, small molecule, or polynucleotide. In some embodiments, the R-spondin-translocation antagonist binds a RSPO1-translocation fusion polypeptide and/or polynucleotide, RSPO2-translocation fusion polypeptide and/or polynucleotide, RSPO3-translocation fusion polypeptide and/or polynucleotide and/or RSPO4-translocation fusion polypeptide and/or polynucleotide. In some embodiments, the R-spondin-translocation antagonist binds a RSPO2-translocation fusion polypeptide and/or polynucleotide. In some embodiments, the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises *EIF3E* and *RSPO2*. In some embodiments, the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises *EIF3E* exon 1 and *RSPO2* exon 2. In some embodiments, the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises *EIF3E* exon 1 and *RSPO2* exon 3. In some embodiments, the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises SEQ ID NO:71. In some embodiments, the R-spondin-translocation fusion polypeptide and/or polynucleotide is a RSPO3-translocation fusion polypeptide and/or polynucleotide. In some embodiments, the RSPO3-translocation fusion polypeptide and/or polynucleotide comprises *PTPRK* and *RSPO3*. In some embodiments, the RSPO3-translocation fusion polypeptide and/or polynucleotide comprises *PTPRK* exon 1 and *RSPO3* exon 2. In some embodiments, the RSPO3-translocation fusion polypeptide and/or polynucleotide comprises *PTPRK* exon 7 and *RSPO3* exon

CLAIMS

WHAT IS CLAIMED IS:

- 1) Use of an effective amount of an ~~R-spondin~~-antagonist specific for R-spondin in the manufacture of a medicament for treating cancer in an individual identified to have an R-spondin chromosomal translocation, wherein treatment is based upon the individual having cancer comprising an R-spondin chromosomal translocation.
- 2) Use of an effective amount of an ~~R-spondin~~-antagonist specific for R-spondin in the manufacture of a medicament for treating a cancer cell identified to have an R-spondin chromosomal translocation *in vivo*, wherein the cancer cell comprises an R-spondin chromosomal translocation.
- 3) Use of an effective amount of an ~~R-spondin~~-antagonist specific for R-spondin in the manufacture of a medicament for treating cancer in an individual provided that the individual has been found to have cancer comprising an R-spondin chromosomal translocation.
- 4) Use of an effective amount of an anti-cancer therapy comprising an ~~R-spondin~~-antagonist specific for R-spondin in the manufacture of a medicament for treating cancer in an individual, wherein a sample obtained from the individual has been determined to comprise an R-spondin chromosomal translocation, whereby the cancer is treated.
- 5) Use of an effective amount of an ~~R-spondin~~-antagonist specific for R-spondin in the manufacture of a medicament for treating cancer, wherein the medicament is to be administered to an individual having cancer that has been identified to have an R-spondin chromosomal translocation and has been selected, wherein the cancer comprises an R-spondin chromosomal translocation, whereby the cancer is treated.
- 6) A method of identifying an individual with cancer who is more or less likely to exhibit benefit from treatment with an anti-cancer therapy comprising an ~~R-spondin~~-antagonist specific for R-spondin, the method comprising: determining presence or absence of an R-spondin chromosomal translocation in a sample obtained from the individual, wherein presence of the R-spondin chromosomal translocation in the sample indicates that the individual is more likely to exhibit benefit from treatment with the anti-cancer therapy comprising the ~~R-spondin~~-antagonist specific for R-spondin or absence of the R-spondin chromosomal translocation indicates that the individual is less likely to exhibit benefit from treatment with the anti-cancer therapy comprising the ~~R-spondin~~-antagonist specific for R-spondin.
- 7) A method for predicting whether an individual with cancer is more or less likely to respond effectively to treatment with an anti-cancer therapy comprising an ~~R-spondin~~-antagonist specific for R-spondin, the method comprising determining an R-spondin chromosomal translocation, whereby presence of the R-spondin chromosomal translocation indicates that the individual is more likely to respond effectively to treatment with the ~~R-spondin~~-antagonist specific for R-spondin and absence of the R-spondin chromosomal translocation indicates that the individual is less likely to respond effectively to treatment with the ~~R-spondin~~-antagonist specific for R-spondin.
- 8) A method of predicting the response or lack of response of an individual with cancer to an anti-cancer therapy comprising an ~~R-spondin~~-antagonist specific for R-spondin comprising detecting in a sample obtained from the individual presence or absence of an R-spondin chromosomal translocation, wherein presence of the R-spondin chromosomal translocation is predictive of response of the individual to the anti-cancer therapy comprising the ~~R-spondin~~-antagonist specific for R-spondin and absence of the R-spondin chromosomal translocation is predictive of lack of response of the individual to the anti-cancer therapy comprising the ~~R-spondin~~ antagonist specific for R-spondin.

~~9)~~ The method of any one of claims ~~6-8~~, wherein the method further comprises administering to the individual an effective amount of an R-spondin antagonist.

~~10)9)~~ An *in vitro* method of inhibiting proliferation of a cancer cell comprising contacting the cancer cell with an effective amount of an R-spondin translocation antagonist specific for R-spondin chromosomal translocation.

~~11)10)~~ Use of an effective amount of an R-spondin translocation antagonist specific for R-spondin chromosomal translocation in the manufacture of a medicament for inhibiting proliferation of a cancer cell *in vivo* wherein said cancer cell is to be contacted with said medicament.

~~12)11)~~ Use of an effective amount of an R-spondin translocation antagonist specific for R-spondin chromosomal translocation in the manufacture of a medicament for treating cancer in an individual.

~~13)12)~~ The method of claim ~~10~~9 or use of claims ~~11~~10 or ~~12~~11, wherein the cancer or cancer cell comprises an R-spondin chromosomal translocation.

~~14)13)~~ The use of any one of claims 1-5 or method of any one of claims ~~6-9~~8 or the method or use of claim ~~13~~12, wherein the R-spondin chromosomal translocation is a RSPO1 translocation, RSPO2 translocation, RSPO3 translocation and/or RSPO4 translocation.

~~15)14)~~ The use or method of claim ~~14~~13, wherein the R-spondin chromosomal translocation is a RSPO2 translocation.

~~16)15)~~ The use or method of claim ~~15~~14, wherein the RSPO2 translocation comprises *EIF3E* and *RSPO2*.

~~17)16)~~ The use or method of claim ~~15~~14, wherein the RSPO2 translocation comprises *EIF3E* exon 1 and *RSPO2* exon 2.

~~18)17)~~ The use or method of claim ~~15~~14, wherein the RSPO2 translocation comprises *EIF3E* exon 1 and *RSPO2* exon 3.

~~19)18)~~ The use or method of claim ~~15~~14, wherein the RSPO2 translocation comprises SEQ ID NO:71.

~~20)19)~~ The use or method of claim ~~14~~13, wherein the R-spondin chromosomal translocation is a RSPO3 translocation.

~~21)20)~~ The use or method of claim ~~20~~19, wherein the RSPO3 translocation comprises *PTPRK* and *RSPO3*.

~~22)21)~~ The use or method of claim ~~20~~19, wherein the RSPO3 translocation comprises *PTPRK* exon 1 and *RSPO3* exon 2.

~~23)22)~~ The use or method of claim ~~20~~19, wherein the RSPO3 translocation comprises *PTPRK* exon 7 and *RSPO3* exon 2.

~~24)23)~~ The use or method of claim ~~20~~19, wherein the RSPO3 translocation comprises SEQ ID NO:72 and/or SEQ ID NO:73.

~~25)24)~~ The use of any one of claims 1-5 or method of any one of claims ~~6-9~~8 or the use or method of any one of claims ~~13~~12-~~24~~23, wherein the R-spondin chromosomal translocation is detected at the chromosomal level (e.g., FISH), DNA level, RNA level (e.g., RSPO1-translocation fusion transcript), and/or protein level (e.g., RSPO1-translocation fusion polypeptide).

~~26)25)~~ The use of any one of claims 1-5 or ~~11~~10-~~12~~11 or method of any one of claims ~~6-10~~9 or the use or method of any one of claims ~~13~~12-~~25~~24, wherein the cancer or cancer cell is colorectal cancer.

~~27)26)~~ The use or method of claim ~~26~~25, wherein the cancer or cancer cell is a colon cancer or rectal cancer.

~~28)27)~~ The use of any one of claims 1-5 or method of any one of claims ~~6-9~~8 or the use or method of any one of claims ~~14~~13-~~27~~26, wherein the R-spondin antagonist specific for R-spondin is an antibody, binding polypeptide, small molecule, or polynucleotide.

~~29)28)~~ The use of any one of claims 1-5 or method of any one of claims ~~6-9~~8 or the use or method of any one of claims ~~14~~13-~~28~~27, wherein the R-spondin antagonist specific for R-spondin is an isolated monoclonal antibody which binds R-spondin.

~~30~~29) The use or method of claim ~~29~~28, wherein the R-spondin is RSPO2 and/or RSPO3.

~~31~~30) The use or method of claim ~~30~~29, wherein the R-spondin is RSPO3.

~~32~~31) The use or method of any one of claims ~~29~~28-~~30~~29, wherein the ~~R-spondin-antagonist specific for R-spondin is~~ an R-spondin-~~chromosomal~~ translocation antagonist.

~~33~~32) An isolated ~~R-spondin-translocation~~ antagonist specific for R-spondin chromosomal translocation, wherein the R-spondin-~~chromosomal~~ translocation antagonist is an antibody, binding polypeptide, small molecule, or polynucleotide.

~~34~~33) The use or method of claim ~~33~~31 or antagonist of claim ~~33~~32, wherein the ~~R-spondin-translocation-antagonist specific for R-spondin chromosomal translocation~~ binds a RSPO1-translocation fusion polypeptide and/or polynucleotide, RSPO2-translocation fusion polypeptide and/or polynucleotide, RSPO3-translocation fusion polypeptide and/or polynucleotide and/or RSPO4-translocation fusion polypeptide and/or polynucleotide.

~~35~~34) The use or method or antagonist of claim ~~34~~33, wherein the ~~R-spondin-translocation-antagonist specific for R-spondin chromosomal translocation~~ binds a RSPO2-translocation fusion polypeptide and/or polynucleotide.

~~36~~35) The use or method or antagonist of claim ~~35~~34, wherein the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises *EIF3E* and *RSPO2*.

~~37~~36) The use or method or antagonist of claim ~~35~~34, wherein the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises *EIF3E* exon 1 and *RSPO2* exon 2.

~~38~~37) The use or method or antagonist of claim ~~35~~34, wherein the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises *EIF3E* exon 1 and *RSPO2* exon 3.

~~39~~38) The use or method or antagonist of claim ~~35~~34, wherein the RSPO2-translocation fusion polypeptide and/or polynucleotide comprises SEQ ID NO:71.

~~40~~39) The use or method or antagonist of claim ~~34~~33, wherein the R-spondin-translocation fusion polypeptide and/or polynucleotide is a RSPO3-translocation fusion polypeptide and/or polynucleotide.

~~41~~40) The use or method or antagonist of claim ~~40~~39, wherein the RSPO3-translocation fusion polypeptide and/or polynucleotide comprises *PTPRK* and *RSPO3*.

~~42~~41) The use or method or antagonist of claim ~~40~~39, wherein the RSPO3-translocation fusion polypeptide and/or polynucleotide comprises *PTPRK* exon 1 and *RSPO3* exon 2.

~~43~~42) The use or method or antagonist of claim ~~40~~39, wherein the RSPO3-translocation fusion polypeptide and/or polynucleotide comprises *PTPRK* exon 7 and *RSPO3* exon 2.

~~44~~43) The use or method or antagonist of claim ~~40~~39, wherein the RSPO3-translocation fusion polypeptide and/or polynucleotide comprises SEQ ID NO:72 and/or SEQ ID NO:73.

~~45~~44) The use of any one of claims 1-5 or ~~11~~10-~~12~~11, or method of any one of claims 6-~~10~~9, or the use or method of any one of claims ~~13~~12-~~32~~31, or the antagonist of claim ~~33~~32, or the use or method or antagonist of any one of claims ~~34~~33-~~44~~43, wherein an additional therapeutic agent is to be administered.