receptor expression has been observed (Brennan & Brown, 2004, *J. Mammary Gland Biol. Neoplasia*, 9:119-31; Malovanovic et al., 2004, *Int. J. Oncol.*, 25:1337-42).

[008] Activation of the Wnt pathway is also associated with colorectal cancer. Approximately 5-10% of all colorectal cancers are hereditary with one of the main forms being familial adenomatous polyposis (FAP), an autosomal dominant disease in which about 80% of affected individuals contain a germline mutation in the adenomatous polyposis coli (APC) gene. Mutations have also been identified in other Wnt pathway components including Axin and β-catenin. Individual adenomas are clonal outgrowths of epithelial cells containing a second inactivated allele, and the large number of FAP adenomas inevitably results in the development of adenocarcinomas through additional mutations in oncogenes and/or tumor suppressor genes. Furthermore, activation of the Wnt signaling pathway, including loss-of-function mutations in APC and stabilizing mutations in β-catenin, can induce hyperplastic development and tumor growth in mouse models (Oshima et al., 1997, *Cancer Res.*, 57:1644-9; Harada et al., 1999, *EMBO J.*, 18:5931-42).

[009] Similar to breast cancer and colon cancer, melanoma often has constitutive activation of the Wnt pathway, as indicated by the nuclear accumulation of β-catenin. Activation of the Wnt/β-catenin pathway in some melanoma tumors and cell lines is due to modifications in pathway components, such as APC, ICAT, LEF1 and β-catenin (see e.g., Larue et al. 2006, *Frontiers Biosci.*, 11:733-742). However, there are conflicting reports in the literature as to the exact role of Wnt/β-catenin signaling in melanoma. For example, one study found that elevated levels of nuclear β-catenin correlated with improved survival from melanoma, and that activated Wnt/β-catenin signaling was associated with decreased cell proliferation (Chien et al., 2009, *PNAS*, 106:1193-1198).

[010] The focus of cancer drug research is shifting toward targeted therapies aimed at genes, proteins, and pathways involved in human cancer. There is a need for new agents targeting signaling pathways and new combinations of agents that target multiple pathways that could provide therapeutic benefit for cancer patients. Thus, biomolecules (e.g., anti-RSPO3 antibodies) that disrupt β-catenin signaling are a potential source of new therapeutic agents for cancer, as well as other β-catenin-associated diseases.

BRIEF SUMMARY OF THE INVENTION

[011] The present ~~invention~~ disclosure provides binding agents, such as antibodies, that bind RSPO3 proteins, as well as compositions, such as pharmaceutical compositions, comprising the binding agents. Binding agents that bind RSPO3 as well as at least one additional antigen or target, and pharmaceutical compositions of such binding agents, are also provided. In certain ~~embodiments~~examples, the RSPO3-binding agents are novel polypeptides, such as antibodies, antibody fragments, and other polypeptides related to such antibodies. The ~~invention~~ present disclosure further provides methods of inhibiting the growth of a tumor by administering the RSPO3-binding agents to a subject with a tumor. The ~~invention~~ present disclosure further provides

methods of treating cancer by administering the RSPO3-binding agents to a subject in need thereof. In some ~~embodiments~~examples, the methods of treating cancer or inhibiting tumor growth comprise targeting cancer stem cells with the RSPO3-binding agents. In some ~~embodiments~~examples, the methods comprise disrupting β-catenin signaling. In some ~~embodiments~~examples, the methods comprise modulating (e.g., inhibiting) angiogenesis. In certain ~~embodiments~~examples, the methods comprise reducing the frequency of cancer stem cells in a tumor, reducing the number of cancer stem cells in a tumor, reducing the tumorigenicity of a tumor, and/or reducing the tumorigenicity of a tumor by reducing the number or frequency of cancer stem cells in the tumor.

[012] ~~In one aspect, the invention provides~~ Disclosed is a binding agent, such as an antibody, that specifically binds human RSPO3. The sequence of human RSPO3 is known in the art and is included herein as SEQ ID NO:3. In certain ~~embodiments~~examples, the RSPO3-binding agent binds within amino acids 22-272 of human RSPO3. In certain ~~embodiments~~examples, the RSPO3-binding agent binds within amino acids 22-207 of human RSPO3. In certain ~~embodiments~~examples, the RSPO3-binding agent binds within amino acids 35-135 of human RSPO3. In certain ~~embodiments~~examples, the RSPO3-binding agent binds within amino acids 35-86 of human RSPO3. In certain ~~embodiments~~examples, the RSPO3-binding agent binds within amino acids 92-135 of human RSPO3. In some ~~embodiments~~examples, the RSPO3-binding agent (e.g., an antibody) specifically binds at least one other human RSPO selected from the group consisting of RSPO1, RSPO2, and RSPO4. In some ~~embodiments~~examples, the RSPO3-binding agent or antibody modulates β-catenin activity, is an antagonist of β-catenin signaling, inhibits β-catenin signaling, and/or inhibits activation of β-catenin. In some ~~embodiments~~examples, the RSPO3-binding agent inhibits RSPO3 signaling. In some ~~embodiments~~examples, the RSPO3-binding agent inhibits, interferes with, and/or disrupts binding of RSPO3 to one or more LGR proteins (e.g., LGR4, LGR5, and/or LGR6). In some ~~embodiments~~examples, the RSPO3-binding agent inhibits binding of RSPO3 to LGR5.

[012a] In one aspect, there is provided an isolated antibody that specifically binds human R-spondin 3 (RSP03), which comprises:

(a) a heavy chain CDR1 comprising DYSIH (SEQ ID NO:78), KASGYTFTDYS (SEQ ID NO:9), or KASGYTFTSYTF (SEQ ID NO:34), a heavy chain CDR2 comprising YIYPSNGDSGYNQKFK (SQ ID NO:79) or IYPSNGDS (SEQ ID NO: 10), and a heavy chain CDR3 comprising TYFANNFD (SEQ ID NO:80), ATYFANYFDY (SEQ ID NO:11), or ATYFANNFDY (SEQ ID NO:35); and

(b) a light chain CDR1 comprising KASQSVDYDGDSYMN (SEQ ID NO:81) or QSVDYDGDSYM (SEQ ID NO:12), a light chain CDR2 comprising AASNLES (SEQ ID NO:82) or AAS (SEQ ID NO:13), and a light chain CDR3 comprising QQSNEDPLTF (SEQ ID NO:83) or QQSNEDPLT (SEQ ID NO:14).

[013] In certain ~~embodiments~~examples, the RSPO3-binding agent is an antibody which binds human RSPO3. In some ~~embodiments~~examples, the antibody binds human RSPO3 and mouse RSPO3. In

certain ~~embodiments~~examples, the antibody comprises a heavy chain CDR1 comprising KASGYTFTDYS (SEQ ID NO:9), KASGYTFTSYTF (SEQ ID NO:34), or DYSIH (SEQ ID NO:78), a heavy chain CDR2 comprising IYPSNGDS (SEQ ID NO:10) or YIYPSNGDSGYNQKFK (SEQ ID NO:79), and a heavy chain CDR3 comprising ATYFANYFDY (SEQ ID NO:11), ATYFANNFDY (SEQ ID NO:35), or TYFANNFD (SEQ ID NO:80). In some ~~embodiments~~examples, the antibody further comprises a light chain CDR1 comprising QSVDYDGDSYM (SEQ ID NO:12) or KASQSVDYDGDSYMN (SEQ ID NO:81), a light chain CDR2 comprising AAS (SEQ ID NO:13) or AASNLES (SEQ ID NO:82), and a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14) or QQSNEDPLTF (SEQ ID NO:83). In some ~~embodiments~~examples, the antibody comprises a heavy chain CDR1 comprising KASGYTFTDYS (SEQ ID NO:9), a heavy chain CDR2 comprising IYPSNGDS (SEQ ID NO:10), and a heavy chain CDR3 comprising ATYFANNFDY

(SEQ ID NO:35), and/or a light chain CDR1 comprising QSVDYDGDSYM (SEQ ID NO:12), a light chain CDR2 comprising AAS (SEQ ID NO:13), and a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14). In some ~~embodiments~~examples, the antibody comprises a heavy chain CDR1 comprising DYSIH (SEQ ID NO:78), a heavy chain CDR2 comprising YIYPSNGDSGYNQKFK (SEQ ID NO:79), and a heavy chain CDR3 comprising TYFANNFD (SEQ ID NO:80), and/or a light chain CDR1 comprising KASQSVDYDGDSYMN (SEQ ID NO:81), a light chain CDR2 comprising AASNLES (SEQ ID NO:82), and a light chain CDR3 comprising QQSNEDPLTF (SEQ ID NO:83). In some ~~embodiments~~ examples, the antibody comprises a heavy chain CDR1 comprising DYSIH (SEQ ID NO:78), a heavy chain CDR2 comprising YIYPSNGDSGYNQKFK (SEQ ID NO:79), and a heavy chain CDR3 comprising TYFANNFD (SEQ ID NO:80), and/or a light chain CDR1 comprising KASQSVDYDGDSYMN (SEQ ID NO:81), a light chain CDR2 comprising AASNLES (SEQ ID NO:82), and a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14). In some ~~embodiments~~examples, the antibody comprises a heavy chain CDR1 comprising KASGYTFTDYS (SEQ ID NO:9) or DYSIH (SEQ ID NO:78), a heavy chain CDR2 comprising IYPSNGDS (SEQ ID NO:10), and a heavy chain CDR3 comprising TYFANNFD (SEQ ID NO:80), and/or a light chain CDR1 comprising QSVDYDGDSYM (SEQ ID NO:12), a light chain CDR2 comprising AAS (SEQ ID NO:13), and a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14).

[014] In certain ~~embodiments~~examples, the RSPO3-binding agent is an antibody which comprises: (a) a heavy chain CDR1 comprising KASGYTFTDYS (SEQ ID NO:9), KASGYTFTSYTF (SEQ ID NO:34), DYSIH (SEQ ID NO:78), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (b) a heavy chain CDR2 comprising IYPSNGDS (SEQ ID NO:10), YIYPSNGDSGYNQKFK (SEQ ID NO:79), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (c) a heavy chain CDR3 comprising ATYFANYFDY (SEQ ID NO:11), ATYFANNFDY (SEQ ID NO:35), TYFANNFD (SEQ ID NO:80), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (d) a light chain CDR1 comprising QSVDYDGDSYM (SEQ ID NO:12), KASQSVDYDGDSYMN (SEQ ID NO:81), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; (e) a light chain CDR2 comprising AAS (SEQ ID NO:13), AASNLES (SEQ ID NO:82), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions; and (f) a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14), QQSNEDPLTF (SEQ ID NO:83), or a variant thereof comprising 1, 2, 3, or 4 amino acid substitutions. In some ~~embodiments~~examples, the amino acid substitutions are conservative amino acid substitutions. In some ~~embodiments~~examples, the substitutions are made as part of a germline humanization process.

[015] In certain ~~embodiments~~examples, the RSPO3-binding agent is an antibody which comprises: (a) a heavy chain variable region having at least 80% sequence identity to SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:44, SEQ ID NO:45, or SEQ ID NO:62; and/or (b) a light chain variable region having at least 80% sequence identity to SEQ ID NO:17, SEQ ID NO:72, or SEQ ID

NO:86. In certain ~~embodiments~~examples, the RSPO3-binding agent is an antibody that comprises: (a) a heavy chain variable region having at least 90% sequence identity to SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:44, SEQ ID NO:45, or SEQ ID NO:62; and/or (b) a light chain variable region having at least 90% sequence identity to SEQ ID NO:17, SEQ ID NO:72, or SEQ ID NO:86.

[016] In some ~~embodiments~~examples, the RSPO3-binding agent is a monoclonal antibody. In some ~~embodiments~~examples, the monoclonal antibody is an IgG1 antibody. In some ~~embodiments~~examples, the monoclonal antibody is an IgG2 antibody. In some ~~embodiments~~examples, the RSPO3-binding agent is monoclonal antibody 131R002 or monoclonal antibody 131R003. In some ~~embodiments~~examples, the RSPO3-binding agent is an affinity-matured variant of monoclonal antibody 131R002 or monoclonal antibody 131R003. In some ~~embodiments~~examples, the RSPO3-binding agent is a chimeric antibody comprising the antigen-binding sites from antibody 131R002 or antibody 131R003. In some ~~embodiments~~examples, the RSPO3-binding agent is a humanized form of antibody 131R002 or antibody 131R003. In some ~~embodiments~~examples, the RSPO3-binding agent is antibody h131R006A, h131R006B, h131R005/131R007, h131R008, h131R010, or h131R011.

[016a] In another aspect, there is provided a monoclonal antibody that competes for specific binding to RSPO3 with the antibody as described herein.

[017] ~~In another aspect, the invention provides~~ Also disclosed is a binding agent (e.g., an antibody) that competes for specific binding to human RSPO3 with an antibody of the ~~invention~~ present disclosure. In some ~~embodiments~~examples, the binding agent (e.g., an antibody) competes for specific binding to human RSPO3 with an antibody that comprises a heavy chain variable region comprising SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:44, SEQ ID NO:45, or SEQ ID NO:62, and a light chain variable region comprising SEQ ID NO:17, SEQ ID NO:72, or SEQ ID NO:86. In some ~~embodiments~~examples, the antibody with which the RSPO3-binding agent competes is antibody 131R002 or antibody 131R003. In some ~~embodiments~~examples, the antibody with which the RSPO3-binding agent competes is a humanized form of antibody 131R002 or antibody 131R003. In some ~~embodiments~~examples, the antibody with which the RSPO3-binding agent competes is antibody h131R006A, h131R006B, h131R005/131R007, h131R008, h131R010, or h131R011. In some ~~embodiments~~examples, the binding agent competes for specific binding to RSPO3 with an antibody of the invention in an *in vitro* competitive binding assay.

[018] In certain ~~embodiments~~examples, the binding agent is an antibody that binds the same epitope, or essentially the same epitope, on RSPO3 as an antibody of the invention (e.g., 131R002, 131R003, or humanized forms/variants thereof). In certain ~~embodiments~~examples, the binding agent is an antibody that antibody binds the same epitope, or essentially the same epitope, on RSPO3 as antibody h131R005/131R007, h131R008, h131R010, or h131R011.

[019] ~~In still another aspect~~In one example, the binding agent is an antibody that binds an epitope on RSPO3 that overlaps with the epitope on RSPO3 bound by an antibody of the ~~invention~~ present disclosure (e.g., 131R002, 131R003, or humanized forms/variants thereof). In some ~~embodiments~~examples, the binding agent is an antibody that binds an

epitope on RSPO3 that overlaps with the epitope on RSPO3 bound by antibody h131R005/131R007, h131R008, h131R010, or h131R011.

[020] In certain ~~embodiments~~examples of each of the aforementioned ~~aspects or embodiments~~ examples, as well as other aspects and/or embodiments described elsewhere herein, the binding agent is a bispecific antibody. In some ~~embodiments~~examples, the bispecific antibody specifically binds human RSPO3 and a second target. In some ~~embodiments~~examples, the bispecific antibody specifically binds human RSPO3 and human RSPO1. In some ~~embodiments~~examples, the bispecific antibody specifically binds human RSPO3 and human RSPO2. In some ~~embodiments~~examples, the bispecific antibody specifically binds human RSPO3 and human RSPO4. In some ~~embodiments~~examples, the bispecific antibody modulates β-catenin activity. In certain ~~embodiments~~examples, the bispecific antibody inhibits β-catenin activity. In certain ~~embodiments~~examples, the bispecific antibody inhibits β-catenin signaling. In certain ~~embodiments~~examples, the bispecific antibody inhibits activation of β-catenin. In some ~~embodiments~~examples, the bispecific antibody reduces the number of frequency of cancer stem cells. In certain ~~embodiments~~examples, the bispecific antibody comprises two identical light chains. In certain ~~embodiments~~examples, the bispecific antibody is an IgG antibody. In certain ~~embodiments~~examples, the bispecific antibody is an IgG1 antibody. In certain ~~embodiments~~examples, the bispecific antibody is an IgG2 antibody.

[021] In some ~~embodiments~~examples, the bispecific antibody comprises: a first antigen-binding site that specifically binds human RSPO3, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising KASGYTFTDYS (SEQ ID NO:9), KASGYTFTSYTF (SEQ ID NO:34), or DYSIH (SEQ ID NO:78), a heavy chain CDR2 comprising IYPSNGDS (SEQ ID NO:10) or YIYPSNGDSGYNQKFK (SEQ ID NO:79), and a heavy chain CDR3 comprising ATYFANYFDY (SEQ ID NO:11), ATYFANNFDY (SEQ ID NO:35), or TYFANNFD (SEQ ID NO:80). In some ~~embodiments~~examples, the first antigen-binding site comprises a light chain CDR1 comprising QSVDYDGDSYM (SEQ ID NO:12) or KASQSVDYDGDSYMN (SEQ ID NO:81), a light chain CDR2 comprising AAS (SEQ ID NO:13) or AASNLES (SEQ ID NO:82), and a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14) or QQSNEDPLTF (SEQ ID NO:83). In some ~~embodiments~~examples, the bispecific antibody further comprises a second antigen-binding site that specifically binds human RSPO1. In some ~~embodiments~~examples, the bispecific antibody further comprises a second antigen-binding site that specifically binds human RSPO2. Non-limiting examples of antibodies to RSPO1 or antibodies to RSPO2 have been described in, for example, International Patent Application Pub. No. WO 2013/012747. In some ~~embodiments~~examples, the first and second binding sites comprise a common (e.g., identical) light chain.

[022] In some ~~embodiments~~examples, the bispecific antibody comprises: a) a first antigen-binding site that specifically binds human RSPO3, and b) a second antigen-binding site that specifically binds human

RSPO1, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising KASGYTFTDYS (SEQ ID NO:9), KASGYTFTSYTF (SEQ ID NO:34), or DYSIH (SEQ ID NO:78), a heavy chain CDR2 comprising IYPSNGDS (SEQ ID NO:10) or YIYPSNGDSGYNQKFK (SEQ ID

NO:79), and a heavy chain CDR3 comprising ATYFANYFDY (SEQ ID NO:11), ATYFANNFDY (SEQ ID NO:35), or TYFANNFD (SEQ ID NO:80). In some ~~embodiments~~examples, the bispecific antibody comprises: a) a first antigen-binding site that specifically binds human RSPO3, and b) a second antigen-binding site that specifically binds human RSPO2, wherein the first antigen-binding site comprises a heavy chain CDR1 comprising KASGYTFTDYS (SEQ ID NO:9), KASGYTFTSYTF (SEQ ID NO:34), or DYSIH (SEQ ID NO:78), a heavy chain CDR2 comprising IYPSNGDS (SEQ ID NO:10) or YIYPSNGDSGYNQKFK (SEQ ID NO:79), and a heavy chain CDR3 comprising ATYFANYFDY (SEQ ID NO:11), ATYFANNFDY (SEQ ID NO:35), or TYFANNFD (SEQ ID NO:80). In some ~~embodiments~~examples, the first antigen-binding site comprises a light chain CDR1 comprising QSVDYDGDSYM (SEQ ID NO:12) or KASQSVDYDGDSYMN (SEQ ID NO:81), a light chain CDR2 comprising AAS (SEQ ID NO:13) or AASNLES (SEQ ID NO:82), and a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14) or QQSNEDPLTF (SEQ ID NO:83).

[22a] In another aspect, there is provided an isolated monoclonal antibody that specifically binds human RSPO3, which comprises:

(a) a heavy chain variable region having at least 90% sequence identity to SEQ ID NO:44, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:45, or SEQ ID NO:62; and

(b) a light chain variable region having at least 90% sequence identity to SEQ ID NO:86, SEQ ID NO:17, or SEQ ID NO:72.

[023] In some ~~embodiments~~examples, the bispecific antibody specifically binds human RSPO3 and comprises: a heavy chain variable region having at least 90% sequence identity to SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:44, SEQ ID NO:45, or SEQ ID NO:62. In some ~~embodiments~~examples, the bispecific antibody specifically binds human RSPO3 and comprises: a heavy chain variable region having at least 95% sequence identity to SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:44, SEQ ID NO:45, or SEQ ID NO:62. In some ~~embodiments~~examples, the bispecific antibody comprises a first and second binding site, wherein the first and second binding sites comprise a common (e.g., identical) light chain. In some ~~embodiments~~examples, the bispecific antibody comprises a light chain variable region having at least 95% sequence identity to SEQ ID NO:17, SEQ ID NO:72, or SEQ ID NO:86.

[024] In certain ~~embodiments~~examples of each of the aforementioned aspects, as well as other aspects and/or ~~embodiments~~examples described elsewhere herein, the RSPO3-binding agent or antibody is isolated. In some ~~embodiments~~examples, the RSPO3-binding agent or antibody is substantially pure.

[025] ~~In another aspect, the invention provides~~ Also disclosed are polypeptides. In some ~~embodiments~~examples, the polypeptide comprises a sequence selected from the group consisting of: SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID

NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38, SEQ ID NO:39, SEQ ID NO:41, SEQ ID NO:42, SEQ ID NO:44, SEQ ID NO:45, SEQ ID NO:46, SEQ ID NO:47, SEQ ID NO:48, SEQ ID NO:49, SEQ ID NO:62, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:68, SEQ ID NO:69, SEQ ID NO:72, SEQ ID NO:73, SEQ ID NO:74, SEQ ID NO:86, SEQ ID NO:87, and SEQ ID NO:88. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:15 and/or SEQ ID NO:17. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:16 and/or SEQ ID NO:17. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:36 and/or SEQ ID NO:17. In some ~~embodiments~~examples, the polypeptide comprises SEQ

ID NO:37 and/or SEQ ID NO:17. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:44 and/or SEQ ID NO:17. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:45 and/or SEQ ID NO:17. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:62 and/or SEQ ID NO:17. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:44 and/or SEQ ID NO:72. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:45 and/or SEQ ID NO:72. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:62 and/or SEQ ID NO:72. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:44 and/or SEQ ID NO:86. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:45 and/or SEQ ID NO:86. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:62 and/or SEQ ID NO:86.

[026] In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:21 and/or SEQ ID NO:23. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:22 and/or SEQ ID NO:23. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:38 and/or SEQ ID NO:23. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:41 and/or SEQ ID NO:23. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:46 and/or SEQ ID NO:23. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:47 and/or SEQ ID NO:23. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:63 and/or SEQ ID NO:23. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:68 and/or SEQ ID NO:23. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:46 and/or SEQ ID NO:73. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:47 and/or SEQ ID NO:73. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:63 and/or SEQ ID NO:73. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:68 and/or SEQ ID NO:73. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:46 and/or SEQ ID NO:87. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:47 and/or SEQ ID NO:87. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:63 and/or SEQ ID NO:87. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:68 and/or SEQ ID NO:87.

[027] In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:27 and/or SEQ ID NO:29. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:28 and/or SEQ ID NO:29. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:39 and/or SEQ ID NO:29. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:42 and/or SEQ ID NO:29. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:48 and/or SEQ ID NO:29. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:49 and/or SEQ ID NO:29. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:64 and/or SEQ ID NO:29. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:69 and/or SEQ ID

NO:29. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:48 and/or SEQ ID NO:74. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:49 and/or SEQ ID NO:74. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:64 and/or SEQ ID NO:74. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:69 and/or SEQ ID NO:74. In some ~~embodiments~~examples, the polypeptide

comprises SEQ ID NO:48 and/or SEQ ID NO:88. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:49 and/or SEQ ID NO:88. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:64 and/or SEQ ID NO:88. In some ~~embodiments~~examples, the polypeptide comprises SEQ ID NO:69 and/or SEQ ID NO:88.

[028] In some ~~embodiments~~examples, the polypeptide is isolated. In certain ~~embodiments~~examples, the polypeptide is substantially pure. In certain ~~embodiments~~examples, the polypeptide is an antibody or part of any antibody, such as an antibody fragment.

[028a] In another aspect, there is provided an antibody comprising the heavy chain variable region encoded by the plasmid deposited with ATCC as PTA-120420 and the light chain variable region encoded by the plasmid deposited with ATCC as PTA-120421.

[028b] In another aspect, there is provided an isolated polynucleotide molecule comprising a polynucleotide that encodes an antibody as described herein.

[029] ~~In another aspect, the invention provides~~ Also disclosed are isolated polynucleotide molecules comprising a polynucleotide that encodes the antibodies and/or polypeptides of each of the aforementioned aspects, as well as other aspects and/or ~~embodiments~~examples described herein. In some ~~embodiments~~examples, the polynucleotide comprises a sequence selected from the group consisting of SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:24, SEQ ID NO:25, SEQ ID NO:26, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:40, SEQ ID NO:43, SEQ ID NO:50, SEQ ID NO:51, SEQ ID NO:52, SEQ ID NO:53, SEQ ID NO:54, SEQ ID NO:55, SEQ ID NO:65, SEQ ID NO:66, SEQ ID NO:67, SEQ ID NO:70, SEQ ID NO:71, SEQ ID NO:75, SEQ ID NO:76, SEQ ID NO:77, SEQ ID NO:84, SEQ ID NO:85, SEQ ID NO:89, SEQ ID NO:90, SEQ ID NO:91, SEQ ID NO:92, SEQ ID NO:93, SEQ ID NO:94, and SEQ ID NO:95. In some ~~embodiments~~examples, the polynucleotide comprises a polynucleotide that encodes a polypeptide selected from the group consisting of: SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38, SEQ ID NO:39, SEQ ID NO:41, SEQ ID NO:42, SEQ ID NO:44, SEQ ID NO:45, SEQ ID NO:46, SEQ ID NO:47, SEQ ID NO:48, SEQ ID NO:49, SEQ ID NO:62, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:68, SEQ ID NO:69, SEQ ID NO:72, SEQ ID NO:73, SEQ ID NO:74, SEQ ID NO:86, SEQ ID NO:87, and SEQ ID NO:88.

[029a] In another aspect, there is provided an isolated cell comprising (a) the antibody according to as described herein; or (b) the polynucleotide as described herein.

[030] ~~The invention further provides~~ Also disclosed are expression vectors that comprise the polynucleotides, as well as cells that comprise the expression vectors and/or the polynucleotides. In some ~~embodiments~~examples, the cell is a hybridoma cell line. In some ~~embodiments~~examples, the cell is a

monoclonal cell line. In some ~~embodiments~~examples, the cell is a prokaryotic cell. In some ~~embodiments~~examples, the cell is an eukaryotic cell.

[030a] In another aspect, there is provided use of the antibody as described herein in the manufacture of a medicament for inhibiting tumor growth.

[031] ~~In other aspects, the invention provides~~ Also disclosed are methods of inhibiting growth of a tumor, comprising contacting the tumor with an effective amount of a RSPO3-binding agent or antibody, including each of those described herein.

[031a] In another aspect, there is provided use of the antibody as described herein in the manufacture of a medicament for inducing differentiation of tumor cells.

[032] ~~In another aspect, the invention provides~~ Also disclosed are a method of inhibiting the growth of a tumor in a subject, comprising administering to the subject a therapeutically effective amount of a RSPO3-binding agent or antibody, including each of those described herein.

[032a] In yet another aspect, there is provided use of the antibody as described herein in the manufacture of a medicament for inhibiting β-catenin signaling in a cell.

[033] ~~In another aspect, the invention provides~~ Also disclosed are a method of inhibiting β-catenin signaling in a cell, comprising contacting the cell with an effective amount of a RSPO3-binding agent or antibody, including

each of those described herein. In some ~~embodiments~~examples, the cell is a tumor cell. In some ~~embodiments~~examples, the tumor is a colorectal tumor. In some ~~embodiments~~examples, the tumor is an ovarian tumor. In some ~~embodiments~~examples, the tumor is a pancreatic tumor. In some ~~embodiments~~examples, the tumor is a lung tumor. In some ~~embodiments~~examples, the tumor is a breast tumor. In some ~~embodiments~~examples, the tumor expresses elevated levels of at least one RSPO protein. In some ~~embodiments~~examples, the tumor expresses elevated levels of RSPO1. In some ~~embodiments~~examples, the tumor expresses elevated levels of RSPO2. In some ~~embodiments~~examples, the tumor expresses elevated levels of RSPO3. In some ~~embodiments~~examples, the tumor expresses a high level of at least one RSPO protein. In some ~~embodiments~~examples, the tumor expresses a high level of RSPO1. In some ~~embodiments~~examples, the tumor expresses a high level of RSPO2. In some ~~embodiments~~examples, the tumor expresses a high level of RSPO3. In certain ~~embodiments~~examples, the RSPO3-binding agent inhibits growth of the tumor, for example, by reducing the number and/or frequency of cancer stem cells in the tumor. In some ~~embodiments~~examples, the tumor contains a RSPO gene fusion. In some ~~embodiments~~examples, the tumor contains a RSPO2 gene fusion. In some ~~embodiments~~examples, the tumor contains a RSPO3 gene fusion.

[033a] In another aspect, there is provided use of the antibody as described herein in the manufacture of a medicament for reducing the frequency of cancer stem cells in a tumor.

[033b] In another aspect, there is provided use of the antibody as described herein in the manufacture of a medicament for treating cancer.

[034] ~~In another aspect, the invention provides~~ Also disclosed are methods of treating cancer in a subject. In some ~~embodiments~~examples, the method comprises administering to a subject a therapeutically effective amount of any of the RSPO3-binding agents or antibodies described above, as well as those described elsewhere herein. In some ~~embodiments~~examples, the cancer is pancreatic cancer. In some ~~embodiments~~examples, the cancer is colorectal cancer. In some ~~embodiments~~examples, the colorectal cancer comprises an inactivating mutation in the adenomatous polyposis coli (APC) gene. In some ~~embodiments~~examples, the colorectal cancer does not comprise an inactivating mutation in the APC gene. In some ~~embodiments~~examples, the colorectal cancer comprises a wild-type APC gene. In some ~~embodiments~~examples, the colorectal cancer comprises a RSPO gene fusion. In some ~~embodiments~~examples, the colorectal cancer comprises a RSPO2 gene fusion. In some ~~embodiments~~examples, the colorectal cancer comprises a RSPO3 gene fusion. In some ~~embodiments~~examples, the cancer is ovarian cancer. In some ~~embodiments~~examples, the cancer is lung cancer. In some ~~embodiments~~examples, the cancer is breast cancer. In some ~~embodiments~~examples, the cancer expresses elevated levels of at least one RSPO protein. In some ~~embodiments~~examples, the cancer is an ovarian cancer that expresses elevated levels of RSPO3. In some ~~embodiments~~examples, the cancer is lung cancer that expresses elevated levels of RSPO3. In some ~~embodiments~~examples, the cancer is

breast cancer that expresses elevated levels of RSPO3. In some ~~embodiments~~examples, the cancer is pancreatic cancer that expresses elevated levels of RSPO3.

[034a] In another aspect, there is provided use of the antibody as described herein in the manufacture of a medicament for treating a disease associated with activation of β-catenin.

[035] ~~In another aspect, the invention provides~~ Also disclosed are methods of treating a disease in a subject wherein the disease is associated with activation of β-catenin, increased β-catenin signaling, and/or aberrant β-catenin signaling, wherein the method comprises administering to the subject a therapeutically effective amount of a RSPO3-binding agent or antibody, including each of those described herein.

[036] In certain ~~embodiments~~examples of each of the aforementioned aspects, as well as other aspects and/or ~~embodiments~~examples described elsewhere herein, the treatment methods further comprise a step of determining the expression level of at least one RSPO protein in the tumor or cancer.

[036a] In another aspect, there is provided an invitro method of identifying a tumor for treatment with an antibody as described herein comprising: determining if the tumor that has an elevated expression or a high expression level of RSPO3.

[037] ~~In another aspect, the invention provides~~ Also disclosed is a method of identifying a human subject or selecting a human subject for treatment with a RSPO3-binding agent or antibody, including but not limited to, each of those described herein. In some ~~embodiments~~examples, the method comprises determining if the subject has a tumor that has an elevated expression level of a specific RSPO (e.g., RSPO3) as compared to the expression of the same RSPO protein in normal tissue or to a pre-determined level of the same RPSO protein. In some ~~embodiments~~examples, the method comprises identifying a subject for treatment or selecting a subject for treatment if the tumor has an elevated level of RSPO expression. In some ~~embodiments~~examples, the method comprises determining if the subject has a tumor that comprises an inactivating mutation in the APC gene. In some ~~embodiments~~examples, the method comprises identifying a subject for treatment or selecting a subject for treatment if the tumor comprises an inactivating mutation in the APC gene. In some ~~embodiments~~examples, the method comprises determining if the subject has a tumor that comprises a RSPO gene fusion (e.g., a RSPO3 gene fusion). In some ~~embodiments~~examples, the method comprises identifying a subject for treatment or selecting a subject for treatment if the tumor comprises a RSPO gene fusion (e.g., a RSPO3 gene fusion).

[038] In certain ~~embodiments~~examples of each of the aforementioned aspects, as well as other aspects and/or ~~embodiments~~examples described elsewhere herein, the treatment methods comprise administering to the subject the RSPO3-binding agent and at least one additional therapeutic agent.

[038a] In yet another aspect, there is provided a pharmaceutical composition comprising the antibody as described herein and a pharmaceutically acceptable carrier.

[039] Pharmaceutical compositions comprising a RSPO3-binding agent or antibody described herein and a pharmaceutically acceptable carrier are further provided, as are cell lines that produce the RSPO3-binding agents. Methods of treating cancer and/or inhibiting tumor growth in a subject (e.g., a human) comprising administering to the subject an effective amount of a pharmaceutical composition comprising the RSPO3-binding agents are also provided.

[040] Where aspects or ~~embodiments~~examples of the ~~invention~~ present disclosure are described in terms of a Markush group or other grouping of alternatives, the present invention encompasses not only the entire group listed as a whole, but also each member of the group individually and all possible subgroups of the main group, and also the main group absent one or more of the group members. The

present invention also envisages the explicit exclusion of one or more of any of the group members in the claimed invention.

AMENDED CLAIMS
received by the International Bureau on 7 April 2014 (07.04.2014)

WHAT IS CLAIMED IS:

1. An isolated antibody that specifically binds human R-spondin 3 (RSPO3), which comprises:

(a) a heavy chain CDR1 comprising DYSIH (SEQ ID NO:78), KASGYTFTDYS (SEQ ID NO:9), or KASGYTFTSYTF (SEQ ID NO:34), a heavy chain CDR2 comprising YIYPSNGDSGYNQKFK (SQ ID NO:79) or IYPSNGDS (SEQ ID NO:10), and a heavy chain CDR3 comprising TYFANNFD (SEQ ID NO:80), ATYFANYFDY (SEQ ID NO:11), or ATYFANNFDY (SEQ ID NO:35); and

(b) a light chain CDR1 comprising KASQSVDYDGDSYMN (SEQ ID NO:81) or QSVDYDGDSYM (SEQ ID NO:12), a light chain CDR2 comprising AASNLES (SEQ ID NO:82) or AAS (SEQ ID NO:13), and a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14) or QQSNEDPLTF (SEQ ID NO:83).

2. The antibody of claim 1, which comprises:

(a) a heavy chain CDR1 comprising DYSIH (SEQ ID NO:78) or KASGYTFTDYS (SEQ ID NO:9), a heavy chain CDR2 comprising YIYPSNGDSGYNQKFK (SQ ID NO:79), and a heavy chain CDR3 comprising TYFANNFD (SEQ ID NO:80); and a light chain CDR1 comprising KASQSVDYDGDSYMN (SEQ ID NO:81), a light chain CDR2 comprising AASNLES (SEQ ID NO:82), and a light chain CDR3 comprising QQSNEDPLTF (SEQ ID NO:83);

(b) a heavy chain CDR1 comprising KASGYTFTDYS (SEQ ID NO:9) or KASGYTFTSYTF (SEQ ID NO:34), a heavy chain CDR2 comprising IYPSNGDS (SEQ ID NO:10), and a heavy chain CDR3 comprising ATYFANYFDY (SEQ ID NO:11) or ATYFANNFDY (SEQ ID NO:35), and a light chain CDR1 comprising QSVDYDGDSYM (SEQ ID NO:12), a light chain CDR2 comprising AAS (SEQ ID NO:13), and a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14); or

(c) a heavy chain CDR1 comprising KASGYTFTDYS (SEQ ID NO:9) or DYSIH (SEQ ID NO:78), a heavy chain CDR2 comprising IYPSNGDS (SEQ ID NO:10), and a heavy chain CDR3 comprising TYFANNFD (SEQ ID NO:80); and a light chain CDR1 comprising QSVDYDGDSYM (SEQ ID NO:12), a light chain CDR2 comprising AAS (SEQ ID NO:13), and a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14).

3. The antibody of claim 1, which comprises:

(a) a heavy chain variable region having at least 90% sequence identity to SEQ ID NO:44, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:45, or SEQ ID NO:62; and

(b) a light chain variable region having at least 90% sequence identity to SEQ ID NO:86, SEQ ID NO:17, or SEQ ID NO:72.

4. The antibody of claim 3, which comprises:

(a) a heavy chain variable region comprising SEQ ID NO:44 and a light chain variable region comprising SEQ ID NO:17, SEQ ID NO:72, or SEQ ID NO:86;

(b) a heavy chain variable region comprising SEQ ID NO:15 and a light chain variable region comprising SEQ ID NO:17 or SEQ ID NO:72;

(c) a heavy chain variable region comprising SEQ ID NO:16 and a light chain variable region comprising SEQ ID NO:17 or SEQ ID NO:72;

(d) a heavy chain variable region comprising SEQ ID NO:36 and a light chain variable region comprising SEQ ID NO:17 or SEQ ID NO:72;

(e) a heavy chain variable region comprising SEQ ID NO:37 and a light chain variable region comprising SEQ ID NO:17 or SEQ ID NO:72;

(f) a heavy chain variable region comprising SEQ ID NO:45 and a light chain variable region comprising SEQ ID NO:17, SEQ ID NO:72, or SEQ ID NO:86; or

(g) a heavy chain variable region comprising SEQ ID NO:62 and a light chain variable region comprising SEQ ID NO:17, SEQ ID NO:72, or SEQ ID NO:86.

5. An isolated antibody that competes with the antibody according to any one of claims 1-4 for specific binding to RSPO3.

6. The antibody according to any one of claims 1-5, which is a recombinant antibody, a monoclonal antibody, a chimeric antibody, a bispecific antibody, a humanized antibody, a human antibody, an IgG1 antibody, an IgG2 antibody, or an antibody fragment comprising an antigen binding site.

7. An antibody comprising the heavy chain variable region encoded by the plasmid deposited with ATCC as PTA-120420 and the light chain variable region encoded by the plasmid deposited with ATCC as PTA-120421.

8. The antibody according to any one of claims 1-7, which inhibits binding of RSPO3 to at least one leucine-rich repeat containing G protein coupled receptor (LGR).

9. The antibody of claim 8, wherein the LGR is selected from the group consisting of LGR5, LGR4, and LGR6.

10. The antibody according to any one of claims 1-9, which
(a) inhibits RSPO3 signaling;
(b) inhibits activation of β-catenin;
(c) inhibits β-catenin signaling;
(d) inhibits tumor growth;
(e) induces expression of differentiation markers in a tumor;
(f) induces cells in a tumor to differentiate; or
(g) reduces the frequency of cancer stem cells in a tumor.

11. A pharmaceutical composition comprising the antibody according to any one of claims 1-7 and a pharmaceutically acceptable carrier.

12. An isolated polynucleotide molecule comprising a polynucleotide that encodes an antibody according to any one of claims 1-7.

13. A cell comprising:
(a) the antibody according to any one of claims 1-7; or
(b) the polynucleotide of claim 12.

14. The use of the antibody according to any one of claims 1-7 for inhibiting tumor growth.

15. The use of the antibody according to any one of claims 1-7 for inducing differentiation of tumor cells.

16. The use of the antibody according to any one of claims 1-7 for reducing the frequency of cancer stem cells in a tumor.

17. The use of the antibody according to any one of claims 1-7 for inhibiting β-catenin signaling in a cell.

18. The use of the antibody of claim 17, wherein the cell is a tumor cell.

19. The use of the antibody according to any one of claims 14-16 or 18, wherein the tumor is a pancreatic tumor, lung tumor, colorectal tumor, or ovarian tumor.

20. The use of the antibody according to any one of claims 1-7 for treating cancer.

21. The use of the antibody of claim 20, wherein the cancer is colorectal cancer, pancreatic cancer, lung cancer, or ovarian cancer.

22. The use of the antibody according to any one of claims 14-16 or 18-21, wherein the tumor or the cancer has a RSPO gene fusion.

23. The use of the antibody according to any one of claims 14-16 or 18-21, further comprising a step of determining if the tumor or cancer has a RSPO gene fusion.

24. The use of the antibody of claim 22 or claim 23, wherein the RSPO gene fusion is a RSPO2 gene fusion or a RSPO3 gene fusion.

25. The use of the antibody according to any one of claims 1-7 for treating a disease associated with activation of β-catenin.

26. The use of the antibody according to any one of claims 14-16 or 18-25, which comprises administering at least one additional therapeutic agent.

27. The use of the antibody of claim 26, wherein the additional therapeutic agent is a chemotherapeutic agent, an angiogenesis inhibitor, an additional antibody.

28. A method of identifying a tumor for treatment with an antibody according to any one of claims 1-7 comprising: determining if the tumor that has a RSPO gene fusion.

29. The method of claim 28, wherein the RSPO gene fusion is a RSPO2 gene fusion or a RSPO3 gene fusion.

AMENDED SHEET (ARTICLE 19)

30. The method according to claim 28 or claim 29, wherein the RSPO gene fusion is determined by a PCR-based assay, microarray analysis, or nucleotide sequencing.

WHAT IS CLAIMED IS:

1. An isolated monoclonal antibody that specifically binds human R-spondin 3 (RSPO3), which comprises:

(a) a heavy chain CDR1 comprising DYSIH (SEQ ID NO:78), KASGYTFTDYS (SEQ ID NO:9), or KASGYTFTSYTF (SEQ ID NO:34), a heavy chain CDR2 comprising YIYPSNGDSGYNQKFK (SQ ID NO:79) or IYPSNGDS (SEQ ID NO:10), and a heavy chain CDR3 comprising TYFANNFD (SEQ ID NO:80), ATYFANYFDY (SEQ ID NO:11), or ATYFANNFDY (SEQ ID NO:35); and

(b) a light chain CDR1 comprising KASQSVDYDGDSYMN (SEQ ID NO:81) or QSVDYDGDSYM (SEQ ID NO:12), a light chain CDR2 comprising AASNLES (SEQ ID NO:82) or AAS (SEQ ID NO:13), and a light chain CDR3 comprising QQSNEDPLTF (SEQ ID NO:83) or QQSNEDPLT (SEQ ID NO:14).

2. The antibody of claim 1, which comprises:

(a) a heavy chain CDR1 comprising DYSIH (SEQ ID NO:78), a heavy chain CDR2 comprising YIYPSNGDSGYNQKFK (SQ ID NO:79), and a heavy chain CDR3 comprising TYFANNFD (SEQ ID NO:80); and

(b) a light chain CDR1 comprising KASQSVDYDGDSYMN (SEQ ID NO:81), a light chain CDR2 comprising AASNLES (SEQ ID NO:82), and a light chain CDR3 comprising QQSNEDPLTF (SEQ ID NO:83).

3. The antibody of claim 1, which comprises:

(a) a heavy chain CDR1 comprising DYSIH (SEQ ID NO:78) or KASGYTFTDYS (SEQ ID NO:9), a heavy chain CDR2 comprising YIYPSNGDSGYNQKFK (SQ ID NO:79), and a heavy chain CDR3 comprising TYFANNFD (SEQ ID NO:80); and a light chain CDR1 comprising KASQSVDYDGDSYMN (SEQ ID NO:81), a light chain CDR2 comprising AASNLES (SEQ ID NO:82), and a light chain CDR3 comprising QQSNEDPLTF (SEQ ID NO:83);

(b) a heavy chain CDR1 comprising KASGYTFTDYS (SEQ ID NO:9) or KASGYTFTSYTF (SEQ ID NO:34), a heavy chain CDR2 comprising IYPSNGDS (SEQ ID NO:10), and a heavy chain CDR3 comprising

ATYFANYFDY (SEQ ID NO:11) or ATYFANNFDY (SEQ ID NO:35), and a light chain CDR1 comprising QSVDYDGDSYM (SEQ ID NO:12), a light chain CDR2 comprising AAS (SEQ ID NO:13), and a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14);

or

(c) a heavy chain CDR1 comprising KASGYTFTDYS (SEQ ID NO:9) or DYSIH (SEQ ID NO:78), a heavy chain CDR2 comprising IYPSNGDS (SEQ ID NO:10), and a heavy chain CDR3 comprising TYFANNFD (SEQ ID NO:80); and a light chain CDR1 comprising QSVDYDGDSYM (SEQ ID NO:12), a light chain CDR2 comprising AAS (SEQ ID NO:13), and a light chain CDR3 comprising QQSNEDPLT (SEQ ID NO:14).

4. An isolated monoclonal antibody that specifically binds human RSPO3, which comprises:

(a) a heavy chain variable region having at least 90% sequence identity to SEQ ID NO:44, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:45, or SEQ ID NO:62; and

(b) a light chain variable region having at least 90% sequence identity to SEQ ID NO:86, SEQ ID NO:17, or SEQ ID NO:72.

5. The antibody of claim 4, which comprises:

(a) a heavy chain variable region comprising SEQ ID NO:44 and a light chain variable region comprising SEQ ID NO:86, SEQ ID NO:72, or SEQ ID NO:17;

(b) a heavy chain variable region comprising SEQ ID NO:15 and a light chain variable region comprising SEQ ID NO:17 or SEQ ID NO:72;

(c) a heavy chain variable region comprising SEQ ID NO:16 and a light chain variable region comprising SEQ ID NO:17 or SEQ ID NO:72;

(d) a heavy chain variable region comprising SEQ ID NO:36 and a light chain variable region comprising SEQ ID NO:17 or SEQ ID NO:72;

(e) a heavy chain variable region comprising SEQ ID NO:37 and a light chain variable region comprising SEQ ID NO:17 or SEQ ID NO:72;

(f) a heavy chain variable region comprising SEQ ID NO:45 and a light chain variable region comprising SEQ ID NO:17, SEQ ID NO:72, or SEQ ID NO:86; or

(g) a heavy chain variable region comprising SEQ ID NO:62 and a light chain variable region comprising SEQ ID NO:17, SEQ ID NO:72, or SEQ ID NO:86.

6. The antibody of claim 5, which comprises a heavy chain variable region comprising SEQ ID NO:44 and a light chain variable region comprising SEQ ID NO:86.

7. A monoclonal antibody that competes for specific binding to RSPO3 with the antibody according to any one of claims 1-6.

8. The antibody according to any one of claims 1-7, which is a recombinant antibody, a chimeric antibody, a bispecific antibody, a humanized antibody, a human antibody, an IgG1 antibody, an IgG2 antibody, or an antibody fragment comprising an antigen binding site.

9. An antibody comprising the heavy chain variable region encoded by the plasmid deposited with ATCC as PTA-120420 and the light chain variable region encoded by the plasmid deposited with ATCC as PTA-120421.

10. The antibody according to any one of claims 1-9, which inhibits binding of RSPO3 to at least one leucine-rich repeat containing G protein coupled receptor (LGR).

11. The antibody of claim 10, wherein the LGR is selected from the group consisting of LGR5, LGR4, and LGR6.

12. The antibody according to any one of claims 1-11, which

(a) inhibits RSPO3 signaling;

(b) inhibits activation of β-catenin;

(c) inhibits β-catenin signaling;

(d) inhibits tumor growth;

(e) induces expression of differentiation markers in a tumor;

(f) induces cells in a tumor to differentiate; or

(g) reduces the frequency of cancer stem cells in a tumor.

13. A pharmaceutical composition comprising the antibody according to any one of claims 1-9 and a pharmaceutically acceptable carrier.

14. An isolated polynucleotide comprising a nucleotide sequence that encodes the antibody according to any one of claims 1-9.

15. An isolated cell comprising:

(a) the antibody according to any one of claims 1-9; or

(b) the polynucleotide of claim 14.

16. Use of the antibody according to any one of claims 1-9, in the manufacture of a medicament for inhibiting tumor growth.

17. Use of the antibody according to any one of claims 1-9, in the manufacture of a medicament for inducing differentiation of tumor cells.

18. Use of the antibody according to any one of claims 1-9, in the manufacture of a medicament for inhibiting β-catenin signaling in a cell.

19. The use of the antibody of claim 18, wherein the cell is a tumor cell.

20. The use of the antibody according to any one of claims 16, 17 or 19, wherein the tumor is a pancreatic tumor, lung tumor, colorectal tumor, or ovarian tumor.

21. The use of the antibody according to any one of claims 1-9, in the manufacture of a medicament for the treatment of cancer.

22. The use of the antibody of claim 21, wherein the cancer is colorectal cancer, pancreatic cancer, lung cancer, or ovarian cancer.

23. The use of the antibody according to any one of claims 16, 17 or 19-22, wherein the tumor or the cancer comprises a RSPO gene fusion.

24. The use of the antibody according to any one of claims 16, 17 or 19-22, wherein the inhibiting tumor growth, inducing differentiation of tumor cells, inhibiting β-catenin signaling in a cell, and treatment of cancer comprises determining if the tumor or cancer comprises a RSPO gene fusion.

25. The use of the antibody according to any one of claims 16, 17 or 19-22, wherein the tumor or the cancer has an elevated expression or a high expression level of RSPO3.

26. The use of the antibody according to any one of claims 16, 17 or 19-22, wherein the inhibiting tumor growth, inducing differentiation of tumor cells, inhibiting β-catenin signaling in a cell, and treatment of cancer comprises determining if the tumor or cancer has an elevated expression or a high expression level of RSPO3.

27. The use of the antibody according to any one of claims 1-9, in the manufacture of a medicament for treating a disease associated with activation of β-catenin.

28. The use of the antibody according to any one of claims 16, 17 or 19-27, wherein the inhibiting tumor growth, inducing differentiation of tumor cells, inhibiting β-catenin signaling in a cell, treatment of cancer, and treating a disease associated with activation of β-catenin comprises administration of the antibody and at least one additional therapeutic agent.

29. The use of the antibody of claim 28, wherein the at least one additional therapeutic agent is a chemotherapeutic agent, an angiogenesis inhibitor, or an additional antibody.

30. An in vitro method of identifying a tumor for treatment with an antibody according to any one of claims 1-9 comprising: determining if the tumor that has an elevated expression or a high expression level of RSPO3.

31. The method according to claim 30, wherein the expression level of RSPO3 is determined by a PCR-based assay, microarray analysis, or nucleotide sequencing.