Abstract. There is no radical cure for all cancer types. The most frequently used therapies are surgical treatment, radiotherapy and chemotherapy. However, recrudescence, radiation resistance and chemotherapy resistance are the most challenging issues in clinical practice. To address these issues, they should be further studied at the molecular level, and the signaling pathways involved represent a promising avenue for this research. In the present review, we mainly discuss the components and mechanisms of activation of the Notch and Wnt signaling pathways, and we summarize the recent research efforts on these two pathways in different cancers. We also evaluate the ideal drugs that could target these two signaling pathways for cancer therapy, summarize alterations in the Notch and Wnt signaling pathways in cancer, and discuss potential signaling inhibitors as effective drugs for cancer therapy.

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1. Introduction

It is well known that cancer is currently one of the leading causes of mortality in the world, and curing it still remains a great challenge to scientists and doctors. Many hypotheses and therapies have been suggested to explain and treat cancer, and these have given hope to physicians and patients because many satisfactory results have been reported. However, there is currently no cancer cure and the mystery of cancer remains unsolved. The mechanism of tumor formation is largely unclear, and present hypotheses cannot explain all of the cases of tumor formation seen in medical practice. Even more noteworthy, therapy seems to be ineffective in some cases, especially in the terminal stages of cancer (1).

Theories regarding cancer biology can guide therapeutic strategies. The role of signal transduction pathways in cancer has been thoroughly investigated. Signaling pathways control the normal development of tissues and organs (2-5), and mutations in signaling molecules may lead to disease, even malignant cancer. A signaling pathway is a group of molecules that transduce signals intracellularly and intercellularly, affecting the fate of cells. The site of signal transduction is important in cancer development, and recent advances in signaling pathway research has provided the means to target therapy to specific sites (5). In addition, better understanding of signaling pathways may help solve the difficult problems of cancer therapy in daily medical practice, such as drug resistance (6). Shi et al (7) reported that inhibitor of p38 MAPK would increase drug sensitivity in colon cancer cell lines. A recent study also found that a decrease in SMAD4 promoter activity may lead to drug resistance in breast cancer cell lines (8). The data indicate better understanding of signaling pathway function may provide insight into the mechanism of drug resistance and reveal potential target sites for improving drug sensitivity. Increasing evidence showed that mutations in signaling pathway components may also be an indicator of prognosis (9). A recent report showed that high Notch1 expression is associated with low survival rate in esophageal squamous cell cancer (10). Furthermore, Chu et al (11) reported that nuclear factor-kappa B also influences the prognosis of oral squamous cell carcinoma. In addition, Wnt5a positivity was reported as a sign of short survival period in NSCLC (12). We can conclude from these data that signaling pathways may be unique prognosis markers for certain cancer types, and may also provide clues for further research on mechanism of cancer prognosis. Taken together, the view of signaling pathways could be a promising theory to explain cancer formation and may help guide therapy for cancer treatment in future.

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Many cancer-related signaling pathways have been reported, such as Notch, Wnt, NF-κB, Ras, JNK, ERK (3,13-16). These pathways have many aspects in common; for example, they all have a role in cellular proliferation, differentiation and survival. Notably, some signal pathways may play double roles in cancer, acting as both oncogene and suppressor, indicating that approach to therapy, based on signaling pathway sites, should be altered according the cancer type. Abnormal expression of signaling pathway components is observed in many cancers, which serves as a reminder that these components have a strong relationship with cancer. In the present review, we mainly focus on the Notch and Wnt signaling pathways to summarize their mechanism of activation, their role in tumorigenesis and their potential as targets for cancer therapy.

2. Notch signaling pathway

The Notch signaling pathway. The Notch signaling pathway is a conserved ligand-receptor signaling pathway in mammals that contains four Notch receptors and five ligands (17). The four receptors, which are named Notch 1, Notch 2, Notch 3 and Notch 4, share a similar structure. Each Notch receptor, Notch 1-4, has 36, 36, 34 and 29 epidermal growth factor (EGF)-like repeats, respectively (18,19). The ligands can be divided into two major groups: the Delta-like ligands (DLL1, DLL3 and DLL4) and Jagged ligands (Jag1 and Jag2) (17). The four Notch receptors and five ligands are transmembrane molecules, meaning that activation of the Notch signaling pathway controls cell fates by interaction of receptors and ligands on the surface of adjacent cells (20). Furthermore, the activation was shown to be regulated via proteolysis by metalloprotease, tumor necrosis factor-α-converting enzyme (TACE) and γ-secretase (21,22). When intercellular activation occurs, the extracellular and intracellular domains of the Notch receptor (ICN) are released by TACE and γ-secretase, respectively. Then, the extracellular domain binds to the ligand on the surface of an adjacent cell, and ICN translocates into the nucleus. ICN contains ankyrin repeats, a RAM domain, a trans-activation domain (TAD), a nuclear localization signal (NLS) and a PEST domain (23). Each domain is necessary for Notch signaling pathway activity. The ICN forms an active transcriptional complex and plays a direct role in regulating the gene expression (23,24) (Fig. 1). From recent study data, the target genes of the Notch signaling pathway, to name a few, are the Hes family, Hey, NF-κB, VEGF and c-myc. All of these Notch target genes were found to be associated with tumorigenesis (25-27).

It has also been shown that the Notch signaling pathway plays an important role in cell differentiation, proliferation and apoptosis (20). Increasing evidence showed that the Notch
signaling pathway can affect the expression of inflammatory cytokines (28,29), vasculogenesis (30,31) and drug resistance (22). Thus, it is reasonable to speculate that disruptions in the Notch signaling pathway may lead to tumorigenesis, as many studies have reported. There is no doubt that a better understanding of the Notch signaling pathway may lead to new insight into cancer therapy and can bring hope to those who treat and suffer from Notch-related tumors.

The relationship between Notch and cancer. As previously discussed, the Notch signal pathway controls many cellular functions, such as differentiation, proliferation and apoptosis. When these functions are disrupted or altered, the outcome may be detrimental. Interestingly, the function of the Notch signaling pathway strongly depends on the cellular context (32). Notch can act both as an oncogene and suppressor gene (32). We mainly summarize the latest studies of the Notch signaling pathway in different tumors, from both oncogene and anti-oncogene points of view.

Notch signaling pathway in hematological malignancy. When discussing Notch-related cancer, human T lymphoblastic leukemias/lymphomas (T-ALL) should be given special attention as it was the first disease reported to be associated with Notch dysfunction in 1991 (33). T-ALL is a malignant tumor that occurs in young people. It has been shown that the intercellular domain of the Notch 1 receptor is the region that is most often mutated in human T-ALL (34,35). A recent study also showed that mutations of the Notch 1 receptor were detected in ~50% of all T-ALL cases (36). From these data, we posit that the Notch 1 receptor has a strong relationship with T-ALL genesis, but how exactly the Notch 1 receptor contributes to T-ALL needs to be clarified.

Studies have demonstrated that the Notch 1 receptor played an important role in T-cell development and cell fate (37,38), and therefore, Notch 1 receptor dysfunction negatively impacts T-cell function. There are three theories to explain how Notch 1 receptor mutation contributes to T-ALL. The most popular theory suggests that changes in the amino acid sequence (such as substitution, insertion and deletion) that encodes the heterodimerization domain lead to the abnormal sensitivity to the ligand (17,32). Another viewpoint suggests that a nonsense or frameshift mutation in the PEST domain, which was detected in 20-25% of T-ALLs, causes T-ALL (17,32). The PEST domain controls the stability of intercellular Notch 1 (ICN1) and this mutation leads to an increase in the concentration of ICN1 (17,36). The third theory implicates a mutation in the juxtamembrane expansion, which increases the activation of the Notch 1 receptor (39). Interestingly and unexpectedly, Notch 1 receptor mutations in T-ALL were associated with a good prognosis in recent studies (40-42). These results indicate that the Notch signaling pathway may be a novel target for the treatment of T-ALL and its mechanism of action in T-ALL should be further studied in detail.

In addition to T-ALL, the Notch signaling pathway is also dysregulated in other hematological malignancies. Chronic lymphocytic leukemia (CLL) is an incurable neoplasm with abnormal B cells. Increasing evidence from recent studies showed that the Notch signaling pathway plays a role in CLL, with the Notch 1 receptor mutant expressed in ~10-20% patients (17,32,43). Genetic studies reveal that the majority of mutations of the Notch 1 receptor are in the PEST domain (44,45). In contrast to what is observed in T-ALL, Notch 1 receptor mutations indicate a poor prognosis in patients with CLL (46,47), and higher frequency of mutation is also observed in chemorefractory CLL (43). In addition to the Notch 1 receptor, other Notch family members also play an important role in hematological malignancy. The Notch 2 receptor is involved in B cell development, which means that mutations in the Notch 2 receptor will likely lead to abnormal formation of B cells and result in further pathologic changes (48). Increasing evidence shows that a Notch 2 receptor mutation can be detected in tumors of B cell origin (49,50).

Notch signaling pathway in solid tumors. In addition to its important role in the hematological malignancy, the Notch signaling pathway also has been shown to be involved in solid tumors, such as breast, lung, gastric and liver cancer. In these solid tumors, the dual role of Notch as an oncogene and suppressor gene is evident. We mainly focus below on the Notch signaling pathway in breast, lung and gastric cancer, aiming at summarizing the latest research advances on the Notch signaling pathway in these diseases.

Breast cancer. Research into the various roles of the Notch signaling pathway has been ongoing for some time. It is known that the Notch signaling pathway plays a crucial role in breast development (20). Evidence showed that the Notch 4 receptor and Notch 3 receptor control normal breast epithelial cells and luminal cells, respectively (51-53). Alteration of the Notch signaling pathway has the potential to cause breast cancer. Of special interest is that the four receptors of the Notch signaling pathway have different roles in breast cancer. The Notch 1 receptor, Notch 3 receptor and Notch 4 receptor have a negative signaling role in breast cancer (54-56). In recent studies, crosstalk of the Notch 1 receptor and other genes (such as Ras, c-myc and JAG1) was proven to contribute the formation of breast cancer (35,54,57). Both the Notch 3 and Notch 4 receptors were found to have the potential to promote transformation (55,56), and inhibition of Notch 3 receptor expression can reduce metastasis of breast cancer to the bone (58). Therefore, the Notch 3 receptor may be a drug target for the treatment of breast cancer. The Notch 1 receptor was associated with poor prognostic outcomes, while the Notch 2 receptor was associated with a good survival rate, as reported by Parr et al (59). They found that overexpression of the Notch 2 receptor can inhibit the tumor growth and promote tumor cell death, indicating that the Notch 2 receptor can be a promising target for further therapy.

Lung cancer. Lung cancer, which is the leading cause of cancer death in the world, can be divided into two types: small cell lung carcinoma (SCLC) and non-small cell lung carcinoma (NSCLC). Research of the Notch signaling pathway has been ongoing for a long time, but its role in lung cancer remains a subject of debate. As we previously mentioned, the role of the Notch signaling pathway in cancer depends on the cellular context, thus, the study of the Notch signaling pathway in lung cancer should be carried out in both SCLC and NSCLC.
In a KrasG12D-driven endogenous NSCLC mouse model, Notch 1 deletion led to a reduction in tumor formation, while Notch 2 receptor deletion led to increased carcinogenesis (60). It was also found that Notch 2 receptor expression was weak in human NSCLC samples, indicating that Notch 2 may play the role of a tumor suppressor in NSCLC (60). Furthermore, in the study by Yang et al (61), it was found that Notch 1 activation may protect the A549 cell line against the anti-tumor effect of pterostilbene. Licciulli et al (62) also reported that the Notch 1 receptor is essential for tumor formation via suppression of p53. In addition, Notch 1 also seemed to be required for resistance to chemotherapy (63,64) and radiotherapy in lung cancer (65). However, Wael et al (66) found that the Notch 1 receptor can significantly induce apoptosis in SCLC and the A549 adenocarcinoma cell line of NSCLC, while its tumor inhibitory function fails in SCC cells of NSCLC. A recent study demonstrated that the Notch 1 receptor can predict prognosis in lung adenocarcinomas: lower Notch 1 receptor expression in a lung adenocarcinoma cell line and patients with positive Notch 1 receptor expression have a longer survival time and a lower rate of recurrence (67). Taken together, before the role of the Notch 1 receptor in lung cancer can be confirmed, further studies are required. Compared with the Notch 1 receptor, the Notch 3 receptor has received less attention, but its role cannot be ignored. Zhou et al (68) detected Notch 3 receptor expression in different types of lung cancer, and they found that Notch 3 expression was high in lung squamous cell carcinoma and adenocarcinoma, but low in small cell carcinoma, compared with the corresponding non-tumor tissue. In addition, the Notch 3 receptor showed its potential ability of predicting the prognosis of patients with NSCLC (69). In this recent report, high levels of the Notch 3 receptor were detected in ~51.1% of cases and were associated with a short survival rate.

Thus, taking all the evidence above into account, we conclude that the Notch signaling pathway is important in lung cancer genesis, progression and prognosis, and it is a promising target for therapy for lung cancer, though its role varies in different types of lung cancer.

Gastric cancer. Gastric cancer (GC) is one of the most common cancers in the world and it is also the leading cause of cancer-related death (70), despite the fact that surgical resection and lymph node dissections are performed. Among the variety of factors that cause gastric cancer, the Notch signaling pathway seems to play an important role. The Notch 1 signaling pathway, in particular, draws great attention in the research of the Notch signaling pathway in GC. At present, Notch 1 seems to function as an oncoprotein in GC, as evidenced by a recent report that Notch 1 contributed to GC progression by inducing COX2 expression (71). Furthermore, Yao et al (72) reported that activation of the Notch 1 receptor can reduce TNFα-induced apoptosis in BGC-823 cells. Interestingly, though their structures are similar, Notch 1 and Notch 2 receptors have different roles in vivo (73). Sun et al (74) reported that expression of Notch 1 in different types of GC varies, indicating that Notch 1 may be a sign of gastric lesions with intestinal-like phenotypes, while the expression of Notch 2 showed a strong relationship with GC formation. However, the role of Notch 2 in GC remains indistinct. With evidence of an oncogenic role (75) and a suppressor role (76), Notch 2 should be further studied to confirm its function in GC.

In addition to the Notch receptors, ligands of the Notch signaling pathway have also received attention in GC. Recently, Piazzi et al (77) reported that the Delta-Like1 (DLL1) controlled the activation of the Notch 1 receptor in GC. Moreover, evidence also showed that DLL1 was the most important ligand for Notch 1 (78), indicating that DLL1 may be a potential target for Notch 1 receptor. Furthermore, another Notch ligand, Delta-Like4 (DLL4), was also reported to be involved in GC progression. Li et al (79) showed that activation of DLL4 may promote tumor proliferation, migration, invasion and tumorigenicity in SGC7901 via overexpression of MMP-2 proenzyme. Sun et al (80) also showed that DLL4 and Jagged1 siRNA gene therapy may greatly reduce the proliferation and invasion of the SGC7901 cell line.

3. Wnt/β-catenin signaling pathway

The Wnt/β-catenin signaling pathway. The Wnt signaling pathway has been a subject of research during the last several decades, and has been shown to function in cell proliferation, growth, cell fate and differentiation (81,82). Mutation of Wnt signaling pathway components causes many diseases, including cancer (82). It is important to understand the Wnt signaling pathway also offers potential benefits for genetic therapy.

The components of the Wnt signaling pathway can be divided into Wnt ligands and Wnt receptors (83). There are 19 Wnt ligands, which all have a cysteine-rich domain, and may activate different types of Wnt signaling pathway by binding specific Wnt signaling receptors (84). In addition, some Wnt ligands are also involved in cancer formation and progression. Wnt1 encodes a number of glycoproteins and was reported as a sign of advanced metastasis for patients with tumors (85). Wnt3a was found to be overexpressed and associated with the level of MMP-9 in colorectal tumor tissue (86). Moreover, a recent report also showed that Wnt3a can promote the proliferation of MCF-7 cells by downregulating β-catenin acetylation (87).

Normally, the Wnt signaling pathway can be categorized as either the canonical Wnt pathway or non-canonical Wnt pathway (88). In the canonical Wnt pathway, β-catenin is the central molecule that controls the on/off ‘switch’ of the Wnt signaling pathway. The Wnt pathway is in the ‘off’ state when Wnt ligands do not bind to any receptors, and β-catenin is released from the cytomembrane. β-catenin is then captured by a protein complex, which is composed of adenomatous polyposis coli (APC), the scaffolding protein Axin, glycogen synthase kinase 3β (GSK3β) and casein kinase 1 (CK1) (88,89). β-catenin is phosphorylated by CK1 and GSK3β and targeted for proteasomal degradation (89), leading to decreased β-catenin concentration, inhibition of nuclear translocation of β-catenin and thus, inhibition of target gene activation (88-90). When the switch is on, a different mechanism unfolds. Wnt ligands bind to the transmembrane Fz receptor and low-density lipoprotein receptor-related proteins (LRP5/6). Then, CK1 and GSK3β are attracted and function as a phosphorylase to LRP5/6. This leads to inacti-
vation of the protein complex, and β-catenin is able to escape degradation, making it possible for it to enter into the nucleus and promote the transcription of target genes (88,91) (Fig. 2).

The non-canonical pathway, which is β-catenin independent, has two modes of activation: the Wnt/Ca2+ pathway and the planar cell polarity (PCP) pathway (92,93). In the Wnt/Ca2+ pathway, Wnt ligands bind to the Frizzled (Fzd) transmembrane receptor and activate a series of proteins that increase the intracellular calcium level, which activates other signal pathways (89,94). The PCP pathway leads to alterations in the cytoskeletal organization, which may influence cellular movement, metastasis and invasion (88,93,95).

The relationship between Wnt and cancer. It has been demonstrated that the Wnt signaling pathway is involved in deciding cell fate, and mutation of Wnt signaling pathway components also showed a strong association with different types of human cancer, such as lung, breast and ovarian cancer (83,96-98). Among these cancers, the Wnt signaling pathway is most involved in hepatocellular carcinoma (HCC) and colorectal cancer (CRC) (99,100). In this section, we mainly discussed the recent research on the Wnt signaling pathway in HCC and CRC, summarizing the effect and potential drug targets of the Wnt signaling pathway in the two cancer types.

Wnt and hepatocellular carcinoma. HCC is the fifth most common cancer type worldwide, according a recent report (101). HCC has a poor prognosis and present treatments are surgical resection, transplantation and chemotherapy (101). The Wnt signaling pathway plays an important role in hepatocellular development, and increasing evidence shows that the Wnt signaling pathway is also involved in liver tumorigenesis (102). A recent report showed that the Wnt signaling pathway has been observed to be activated in at least 1/3 of HCC (103). Hence, effective inhibition of the Wnt signaling pathway may be a potential treatment for HCC. Hou et al (104) reported in their recent research that mesenchymal stem cells (MSCs) were able to secrete Dkk-1 to inhibit proliferation and promote apoptosis of HepG2 cell lines by inhibiting expression of the Wnt signaling pathway. Their findings provided a new potential mechanism of how MSC injection could cure HCC. Zucchini-Pascal et al (105) also found that activation of the Wnt signaling pathway was essential for the epithelial to mesenchymal transition (EMT) in hepatocarcinoma. A study also showed that β-catenin could influence intercellular adhesion by affecting α-catenin and E-cadherin (106). These reports indicated that the Wnt signaling pathway may also be involved in intrahepatic dissemination and distal metastasis of HCC. Furthermore, Singh et al (107) also found that deficiency of β-catenin could inhibit proliferation of liver adenomas in a...
mouse model. In addition to the effect on tumorigenesis and metastasis in liver, activation of the Wnt signaling pathway could also influence chemotherapy resistance in HCC by promoting the overexpression of aldehyde dehydrogenase isoforms (a marker of chemotherapy resistance in cancer), as reported by Calderaro et al (108). It is well known that liver fibrosis is also involved in HCC, especially viral-related liver cancer (88), and increasing evidence also showed that the Wnt signaling pathway may be involved. Cheng et al (109) found that the Wnt signaling pathway was upregulated in activated hepatic stellate cells. In addition, a recent report showed that liver fibrosis could be improved by inhibiting the Wnt signaling pathway (110).

From these data, we may preliminarily conclude that activation of the Wnt signaling pathway plays an important role in liver fibrosis, hepatocellular tumorigenesis and tumor development, and inhibition of the Wnt signaling pathway could strongly inhibit carcinogenesis.

Wnt and colorectal cancer. Colorectal cancer (CRC) is one of the most common cancers and one of the leading causes of cancer-related mortality in the world (111,112). Gaining insight into the genetic changes of CRC is important for both treatment and diagnosis. Among the various genes involved in CRC, the Wnt signaling pathway has attracted much attention (113), as more than half of CRC cases have a β-catenin mutation (114). A recent report showed that the activation of different parts of Wnt signaling pathway may lead to development of one of two types of colorectal neoplasia: serrated or conventional adenoma/polyp (115). Accumulated research also showed that the application of Wnt signaling pathway inhibitors could greatly promote apoptosis and reduce proliferation of CRC cells (116,117). Tumova et al (118) also reported that the use of monensin could inhibit expression of β-catenin in human colorectal carcinoma cells and decrease cell proliferation, indicating that Wnt/β-catenin could be used as gene target for CRC and monensin is a potential drug for therapy. In addition, evidence from clinical practice also showed that CRC patients have a high rate of overexpression of Wnt signaling pathway proteins (119). Voorham et al (120) reported observation of methylated Wnt pathway antagonists from clinical specimens. It is possible that the Wnt signaling pathway could also be used as a potential diagnostic and prognostic biomarker in CRC patients (119,121). Thus, both experimental and clinical data showed that the over activation of the Wnt signaling pathway may be a potential target for CRC. In addition to these studies of the oncogenic role of Wnt in CRC, Abdelmaksoud-Dammak et al (122) found that Wnt5a expression was lower in tumor compared with normal tissue, indicating that Wnt5a may play a role as an antioncogene in CRC. Interestingly, Bauer and colleagues found that the Wnt5a gene could result in proteins of different length, Wnt5a-long and Wnt5a-short. These two genes have inverse functions, with the Wnt5a-long functioning as a suppressor and Wnt5a-short as an oncogene (123).

From these data, we may conclude that Wnt/β-catenin is an oncogenic pathway in CRC, though some Wnt ligands may act as suppressor, as some studies showed. Compounds that could inactivate Wnt/β-catenin could be used as potential drugs for CRC.

Wnt and esophageal squamous cell carcinoma. Reports have shown that ~95% esophageal cancer is esophageal squamous cell cancer (ESCC), with 15% 5-year survival rate (124). At present, studies have found that Wnt signal pathway has played an indispensable role in the development of ESCC, indicating that the components of Wnt signal pathway could be the potential targets for treating ESCC.

Moyes et al (125) have reported that the expression of nuclear activated β-catenin was highly expressed in high grade of dysplasia when compared with that in Barrett's metaplasia. They also found that factitious expression of Wnt signal pathway in mouse oesophagus could cause tissue disorganization (125). Their results indicated that Wnt signal pathway plays an initiative effect in ESCC. In addition, a recent study showed that Wnt 10a, a component of Wnt signal pathway, promotes the invasive and self-renewing ability of ESCC (126), indicating that abnormal expression of Wnt signal pathway could promote the malignant activity of ESCC. From recent studies we may concluded that Wnt signal pathway is important in tumorigenesis and progression of ESCC, targeting components of Wnt signal pathway may make a contribution to treating ESCC both in the initial and the progression period.

Wnt inhibitory factor-1 (WIF1) is one of the most important Wnt inhibitors. A recent study has shown that WIF1 promoter is methylated in ESCC tissues, and re-expression of WIF1 could decrease the transcription activity of β-catenin/TCF and inhibit the cell proliferation and migration (127). Interestingly, Ge et al (128) found that the low expression of WIF1 in ESCC is due to the high expression of Hotair, which is a well-known long non-coding RNA. They reported that Hotair could directly promote histone H3K27 methylation in WIF1 promoter region and activated the Wnt signal pathway. In addition, Liu et al (129) reported that LKB1, a tumor suppressor, could inhibit the Wnt signal pathway through increasing GSK3β activity, causing low expression of β-catenin. Another tumor suppressor, SOX10, was also reported to inhibit the epithelial to mesenchymal transition (EMT) and stemness ability in ESCC cells, by competing with TCF4 to bind β-catenin (130).

From these studies, we may conclude that Wnt signal pathway plays an important role in ESCC. Abnormal expression of Wnt signal pathway may be the initial factor and the stimulative factor in ESCC. Moreover, studies have shown that targeting the components of Wnt signal pathway could inhibit the malignant activity of ESCC cell lines, indicating it is a potential treatment of ESCC.

Potential drug target for cancer therapy. As we have discussed above, both the Notch and Wnt signaling pathways have been proven to have a strong relationship with different types of tumors, playing dual roles of oncogene and suppressor. This means that the Notch and Wnt signaling pathway can be used as promising targets for therapy. In our opinion, the genetic therapy can be designed in three aspects: process or substance that activates genes, expression of the gene itself and mimic of the function of the genes.

Target Notch signaling pathway. The clinical approach to targeting the Notch signal pathway is divided into the use of
Combining DLL4 antibody and other therapies, such as inhibitors of the Notch pathway, has drawn much attention. DLL4, a transmembrane glycoprotein that functions as a ligand for Notch1, plays an important role in Notch-related stem cell self-renewal, and its overexpression has been found in various tumors (79,131,132). Thus, inhibition of DLL4 seems to be a promising treatment to cure cancer. The humanized phage antibody YW152F, which specifically binds to DLL4 receptor on HUVE cells, inhibiting angiogenesis (137) and was proven to be able to inhibit both tumor growth and angiogenesis (134). Monoclonal anti-DLL4 antibody, also known as 21M18, has drawn much attention. It has been reported that it could inhibit anticancer stem cell, anti-angiogenesis and antitumor growth functions (135,136). Recently, MED10629, an investigational human therapeutic antibody, was also reported to be able to inhibit tumor growth by deregulating angiogenesis (133). The use of monoclonal antibodies against murine DLL4 (HMD4-2) in mice was also proven to be able to inhibit both tumor growth and angiogenesis (134). Monoclonal anti-DLL4 antibody, also known as 21M18, has drawn much attention. It has been reported that it could have anticancer stem cell, anti-angiogenesis and antitumor growth functions (135,136). Recently, MED10629, an investigational human therapeutic antibody, was also reported in anti-interaction of DLL4 and Notch1, inhibiting angiogenesis in vivo (137). In addition, studies also showed that the combined use of DLL4 antibody and other therapies, such as radiation treatment and γ-secretase inhibitor, could improve the overall therapeutic effect (138,139). Besides DLL4, other components of the Notch signaling pathway are targeted by antibodies. Aste-Amézaga et al (140) reported on an antibody that could target Notch1 and inhibit the expression of Notch target genes. Sharma et al (141) also showed the antibody against Notch1 could decrease cell proliferation and induce apoptotic cell death. From the data analyzed in the current study, we conclude that the use of an antibody against Notch seems to be a promising anticaner therapeutic strategy. However, antibodies against the Notch receptor should be used cautiously, for it has been reported that the chronic use of these antibodies could lead to vascular neoplasms (142).

Antibodies targeting the Notch pathway. DLL4 plays an important role in Notch-related stem cell self-renewal, and its overexpression has been found in various tumors (79,131,132). Thus, inhibition of DLL4 seems to be a promising treatment to cure cancer. The humanized phage antibody YW152F, which specifically binds to DLL4 receptor on HUVE cells, inhibiting angiogenesis (137) and was proven to be able to inhibit both tumor growth and angiogenesis (134). Monoclonal anti-DLL4 antibody, also known as 21M18, has drawn much attention. It has been reported that it could inhibit anticancer stem cell, anti-angiogenesis and antitumor growth functions (135,136). Recently, MED10629, an investigational human therapeutic antibody, was also reported to be able to inhibit tumor growth by deregulating angiogenesis (133). The use of monoclonal antibodies against murine DLL4 (HMD4-2) in mice was also proven to be able to inhibit both tumor growth and angiogenesis (134). Monoclonal anti-DLL4 antibody, also known as 21M18, has drawn much attention. It has been reported that it could have anticancer stem cell, anti-angiogenesis and antitumor growth functions (135,136). Recently, MED10629, an investigational human therapeutic antibody, was also reported in anti-interaction of DLL4 and Notch1, inhibiting angiogenesis in vivo (137). In addition, studies also showed that the combined use of DLL4 antibody and other therapies, such as radiation treatment and γ-secretase inhibitor, could improve the overall therapeutic effect (138,139). Besides DLL4, other components of the Notch signaling pathway are targeted by antibodies. Aste-Amézaga et al (140) reported on an antibody that could target Notch1 and inhibit the expression of Notch target genes. Sharma et al (141) also showed the antibody against Notch1 could decrease cell proliferation and induce apoptotic cell death. From the data analyzed in the current study, we conclude that the use of an antibody against Notch seems to be a promising anticaner therapeutic strategy. However, antibodies against the Notch receptor should be used cautiously, for it has been reported that the chronic use of these antibodies could lead to vascular neoplasms (142).

**Inhibition of γ-secretase.** γ-secretase is a membrane protein that catalyzes intramembrane proteolysis (5). As previously mentioned, γ-secretase plays an important role in ICN release. Thus, the use of inhibitors of γ-secretase (GSIs) could be a promising therapy for Notch-related cancer. Rosati et al (143) reported that GSI1 (Z-Leu-Leu-Nle-CHO) could induce apoptosis of primary chronic lymphatic leukemia (CLL) through three apoptosis factors. Palagani and colleagues found that the use of GSI IX could effectively inhibit epithelial mesenchymal transition (EMT) in human pancreatic ductal adenocarcinoma (PDAC) (144). Their findings showed a central role of Notch signal pathway in PDAC and the use of GSI IX has shown its potential in treating PDAC. Furthermore, Schott et al (145) reported that the use of GSI (MRK-003) could increase the efficacy of docetaxel in preclinical studies, indicating that the combination use of GSI and chemotherapy could be beneficial for the treatment of cancer. Similarly, another inhibitor, PF03084014, could also enhance the antitumor effect with the combination of fludarabine (146). Recently, Saito et al (147) reported that the use of γ-secretase inhibitors could inhibit the proliferation of glioma tumor-initiating cells. Groeneweg et al (148) reported that the effect of GSI in vitro is dose-dependent and could decrease tumor growth (Table I).

**Target Wnt/β-catenin signaling pathway.** In the Wnt/β-catenin signal pathway, β-catenin has been proven to play a central role in cancer. Thus, inhibitors of β-catenin could be used for cancer treatment (102). Dahmani et al (150) reported in their review that inhibitors of β-catenin could be divided into three types: those affecting the interaction between Wnt ligands and Fzd receptors, those that destroy complex stability and those affecting the activity of β-catenin in the nucleus.

In investigating the interaction between Wnt ligands and receptors, specific therapeutic antibodies have been widely used (150). Evidence has shown that antibodies that bind to Wnt ligands and receptors could inhibit the Wnt/β-catenin signaling pathway (Table II). Fontenot et al (151) reported in their recent research that SFRP2 monoclonal antibody could induce the antitumor effect and inhibit the Wnt/β-catenin signaling pathway in breast models. Wang and colleagues also found a dose-dependent effect of anti-cadherin-17 antibody in suppressing β-catenin in a HCC model (152). Similarly, Gao et al (153) reported that HS20, a human monoclonal

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**Table I. Summary of the inhibitors in Notch signal pathway.**

<table>
<thead>
<tr>
<th>Target</th>
<th>Drug</th>
<th>Model</th>
<th>Effect</th>
<th>Author</th>
<th>Refs.</th>
</tr>
</thead>
<tbody>
<tr>
<td>DLL4</td>
<td>YW152F</td>
<td>HUVE cells</td>
<td>Deregulating angiogenesis</td>
<td>Ridgway et al</td>
<td>(133)</td>
</tr>
<tr>
<td></td>
<td>HMD4-2</td>
<td>Pancreatic cancer</td>
<td>Inhibit tumor growth and angiogenesis</td>
<td>Oishi et al</td>
<td>(134)</td>
</tr>
<tr>
<td></td>
<td>21M18</td>
<td>Colorectal cancer</td>
<td>Anticancer stem cell, anti-angiogenesis and antitumor growth functions</td>
<td>Fischer et al</td>
<td>(136)</td>
</tr>
<tr>
<td>Notch1</td>
<td>MED10629</td>
<td>HUVE cells</td>
<td>Inhibiting angiogenesis</td>
<td>Jenkins et al</td>
<td>(137)</td>
</tr>
<tr>
<td></td>
<td>602.101 (antibody)</td>
<td>Breast cancer</td>
<td>Decrease cell proliferation and induce apoptotic cell death</td>
<td>Sharma et al</td>
<td>(141)</td>
</tr>
<tr>
<td>γ-secretase</td>
<td>GSI IX</td>
<td>Pancreatic cancer</td>
<td>Inhibit EMT</td>
<td>Palagani et al</td>
<td>(144)</td>
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antibody against glypican-3, could disrupt the stability of Wnt3a and glypican-3 and inhibit the Wnt/β-catenin signaling pathway in HCC cells. A recent study reported that a monoclonal antibody, which could target Fzd receptors and prevent their integration with Wnt ligands, has been widely used in treating cancer (89). Recent studies also showed that LRPS/6, closely related membrane receptors for the Wnt signaling pathway, can also be targeted by antibodies for further treatment in cancer (154,155). In addition to antibodies, a number of molecules could also function as Wnt inhibitors. Wnt inhibitory proteins and secreted Fzd-related proteins were the most commonly studied molecules that bind to their targets, and as a result, they inhibit Wnt/β-catenin activity (150). Fzd receptors were also reported to be potential targets for cancer treatment, as soluble Fzd-7 and Fzd-8 were reported to have antitumor effect (150,156,157). IWP2 and Wnt-C59 could inhibit Wnt protein secretion and thus, prevent Wnt signal activation (101). Other molecules, such as flavonoids, monensin and resveratrol, were also shown to have potential antitumor ability via inhibition of the Wnt/β-catenin signaling pathway (118,158,159).

When targeting β-catenin to the destruction complex stability in cytoplasm, the situation seems to be complex. There are two approaches to targeting β-catenin in cytoplasm: the destruction complex stability and β-catenin itself. Axin, as one of the most important components in the destruction complex, has attracted a lot attention recently. Small peptides, such as IWR2 and IWS5, were reported to prevent Axin from degradation and inhibit β-catenin activity (101). Other approaches to increase Axin stability could use a tankyrase inhibitor, such as XAV939, which could prevent the interaction of tankyrases and Axin (160). Parp poly-(ADP-ribose) polymerase (PARP) could promote the ribosylation of Axin, which would cause the degradation of Axin and increase β-catenin levels. The use of a PARP inhibitor could improve the level and stability of Axin, and thus reduce the activation of β-catenin (161). In addition to Axin, CK1α, another member of the destruction complex, is also a potential target for inhibiting β-catenin activity. In a recent study, Park et al (162) found that calotropin could inhibit the Wnt signaling pathway by increasing CK1α protein levels. Their finding is the first to discover a small molecule that could increase CK1α protein, indicating that calotropin could be used as a potential drug for cancer therapy. Additionally, the use of pyrvinium to treat familial adenomatous polyposis by inhibiting the Wnt signaling pathway via activation of CK1α has also been reported (163).

Another approach to targeting the Wnt/β-catenin signal pathway is nuclear, β-catenin and its co-activators are the targets. In the nucleus, the activation of Wnt/β-catenin signaling is mediated by formation of a β-catenin/Lef-Tcf complex (150). Thus, a molecule that can disrupt this complex could be ideal for treating Wnt-related cancer. Wei et al (164) reported that they found three small molecules that could decrease Tcf4/β-catenin binding capability and transcriptional activity in HCC cell lines, indicating that these three molecules could be used as anti-HCC drugs. Recently, a 2,3,6-trisubstituted quinoxaline derivative (GDK-100017) was also reported as an inhibitor of the β-catenin/Lef-Tcf complex, which also enhances radiosensitivity and reduces cell proliferation (165). In addition, lycopene was also reported to inhibit Wnt-Tcf signaling in breast cancer cells (166). Earlier, streponigrin was reported to inhibit the β-catenin/TCF complex binding to DNA, leading to a proliferation inhibitory effect (167). Furthermore, ICG-001 inhibited β-catenin activity in nucleus by disrupting β-catenin/CBP interaction (168) (Fig. 2).

4. Conclusion

In the present review, we mainly focused on the Notch signaling and Wnt signaling pathways, aiming to briefly describe the role of these two signaling pathway in different types of cancer and discussing inhibitors that may be potential anticancer therapies. Both the Notch and Wnt signaling pathways play an important role in maintaining normal cell fate, and these signaling pathways are also involved in a number of cancers. Better understanding of these signaling mechanisms could lead to better cancer therapies. Mutational and abnormal expression of Notch and Wnt signaling pathway components are observed in different types of cancer, as we have discussed in this review. Targeting these disrupted genes potentially reverse cancer damage, in as yet unknown manner. In recent studies, evidence has shown that the combination of signaling inhibitors and traditional therapy (such as radiotherapy and chemotherapy) can treat cancer more effectively compared with single therapy (145,169). However, the biosafety of signaling pathway inhibitors should be closely monitored, as recent studies reported toxicity associated with chronic antibody application in clinical practice (142). Signaling pathways
play an important role in tumorigenesis, tumor development and prognosis, and targeting signaling pathways has the potential to be effective for treating cancer.

References


165. Lee SB, Gong YD, Park YI and Dong MS: 2,3,6-Trisubstituted quinoxaline derivative, a small molecule inhibitor of the Wnt/ beta-catenin signaling pathway, suppresses cell proliferation and enhances radiosensitivity in A549/Wnt2 cells. Biochem Biophys Res Commun 431: 746-752, 2013.


