Topical Issue on Cancer Signaling, Metastasis and Target Therapy **Open Access**

Review Article

Quan Liang, Wei Li, Zhanchao Zhao, Qiang Fu*

Advancement of Wnt signal pathway and the target of breast cancer

DOI 10.1515/biol-2016-0013
Received April 24, 2016; accepted June 6, 2016

Abstract: Wnt/β-catenin signaling has been proved to play an important role in the development and promotion of cancer metastasis. The activation of Wnt signals can lead to duplicating, updating, metastasizing and relapsing. The Wnt signaling pathway is mainly divided into the Wnt/β-catenin pathway and the Wnt/calcium pathway. A better understanding of all the diverse functions of Wnt and their molecular mechanisms has evoked prevailing interest in identifying additional targets related to the Wnt /β-catenin pathways in breast cancer. A number of new target, related to Wnt /β-catenin pathways have been identified in recent years, including NOP14, BKCa channels, Emilin2, WISP, MicroRNAs, NRBP1, TRAF4, and Wntless. In this review, we will introduce the new targets related to the Wnt /β-catenin pathways in breast cancer.

Keywords: Breast cancer, Wnt signal pathway, microRNA

1 Introduction

Breast cancer is the most common cancer and a leading cause cancer-related death in females [1]. Although improvements in understanding the underlying mechanisms of breast cancer and developing new therapeutic approaches have been recently achieved, more than 400,000 women die from breast cancer every year [2]. Wnt/β-catenin signaling has been shown to play an important role in the development and promotion of cancer metastasis [3]. Upon activation, the Wnt signals stabilize and lead to the accumulation of β-catenin. Activated β-catenin dissociates with E-cadherin, dissembling the adherens and activating expression of target genes, most of which show invasion promotion functions [4]. The activation of Wnt signals leads to duplicating, updating, metastasizing and relapsing. The Wnt signaling pathway is mainly divided into the Wnt/β-catenin pathway and the Wnt/calcium pathway. An overview of the Wnt signaling pathway follows.

2 Wnt/β-Catenin Pathway

The TCF/LEF family of DNA-bound transcription factors participates in regulating the gene for β-catenin [5]. Binding of β-catenin to TCF/LEFs can activate or de-repress Wnt target genes [6]. TCF carry a single high motility group (HMG) domain, sufficient for DNA binding and an N-terminal β-catenin binding domain [5]. In addition, HMG can bind to destruction complexes, consisting of proteins including adenomatous polyposis coli (APC), glycogen synthase kinase (GSK)-3β, casein kinase (CK)-1α and β-catenin, which are brought together by the scaffold [7]. Upon binding to the destruction complex, β-catenin causes phosphorylation, followed by ubiquitination and degradation by the proteasome, when Fz receptors are unoccupied by Wnt ligands [8]. Therefore, without Wnt stimulation, cytoplasmic β-catenin levels are kept low by a degradation complex [9]. However, when Wnt binds to its receptors Frizzled and LRP, the destruction complex cannot promote β-catenin signaling [10]. That state allows APC and axin binding to this membrane complex and prevents the breakdown of β-catenin, therefore free unphosphorylated β-catenin can accumulate and translocate to the nucleus where it binds to transcription factors, including T-cell factor (TCF) and LEF-1 [11,12].

***Corresponding author: Qiang Fu,** Department of General Surgery, Tianjin Medical University General Hospital, Tianjin 300052, China, E-mail: liangq01@126.com
Quan Liang, Zhanchao Zhao, Department of General Surgery, Tianjin Medical University General Hospital, Tianjin 300052, China
Wei Li, Department of Ultrasonography, Tianjin Medical University General Hospital, Tianjin 300052, China

3 Wnt/Calcium Pathway

Calcium signaling activity is sufficient to activate calcium sensitive enzymes, including protein kinase C (PKC), calcium dependent kinase II (CamKII) or calcineurin (CaCN) [13]. PLC cleaves phosphatidylinositol-4, 5-bisphosphate (PIP2) into inositol-1, 4, 5- trisphosphate (IP3) and diacylglycerol (DAG) [14]. IP3 binds to IP3 receptors, which release Ca2+ from subcellular stores such as the endoplasmic reticulum (ER) [15]. Activated calcineurin dephosphorylates the transcription factor and nuclear factor of activated T-cells (NFAT), allowing NFAT to translocate into the nucleus where it activates NFAT-responsive genes [16].

Based on a better understanding of all the diverse functions of Wnt and their molecular mechanisms in recent years, additional targets associated with Wnt signals pathway have been discovered. In this review, we make a summary on these new targets. Investigating the protein and gene, which is associated with Wnt signals pathway, will provide a theoretical basis for targeted treatment of breast cancer.

4 NOP14

NOP14 is a stress-responsive gene that is required for 18S rRNA maturation and 40S ribosome production, interacting with PAXIP1, which plays a critical role in maintaining genome stability, condensation of chromatin and progression through mitosis, containing tandem breast cancer carboxyl-terminal domains and regulating multiple aspects of the cellular response to DNA damage, such as cell survival and differentiation [17–22]. Recent studies have suggested that NOP14 may be related to cancer development. In prostate cancer cells, NOP14, a target gene of the polycomb repressive complex, plays a critical role in neoplastic progression [23,24]. Moreover, high levels of NOP14 mRNA and protein were observed in the fibrocystic breast cell line MCF10A; whereas the levels of NOP14 mRNA and protein were low in the four breast cancer cell lines. Strikingly, NOP14 levels contrast with the malignancy of human breast cancer, which is high in atypical ductal hyperplasia (ADH) and primary cancer but low in the advanced breast cancer tissues. Importantly, the investigators discovered that NOP14 could assemble β-catenin on the membranes of breast cancer cells and prevent its nucleus translocation and the following activation. NOP14 increased APC and β-catenin levels, as well as GSK-3β phosphorylation level in breast cancer cells, and inhibited the entry of β-catenin into the nucleus of breast cancer cells. Additionally, in ER-positive breast cancers, NOP14 increase the level of ERα via NRIP1, implied that NOP14 can suppress breast cancer by inhibiting the Wnt/ β-catenin pathways possibly by up-regulating NRIP1 [25]. These findings provide new hope of developing targeted therapies against NOP14 and NRIP1 for breast cancer.

5 BKCa channel

The large conductance of calcium and voltage activated potassium (BKCa) channels function as oncogenes in breast cancers [26]. Through gene amplification, alternative splicing, and increased protein half-life, BKCa channels are overexpressed in many types of cancers [27-30]. In glioma cells, BKCa channels are somewhat more sensitive to calcium and voltage than other BK channels, and thus generate K+ currents in environments where their normal counterparts are silent [27]. Higher grade tumors, characterized by enhanced growth and invasiveness, express more BK channels than lower grade tumors [27]. BKCa channels generate vast amounts of outward K+ currents and therefore are powerful modulators of the transmembrane potential of a cell. The investigators observed that BKCa channels also function as oncogenes in β-catenin-positive breast cancer; they direct their oncogenic input towards sustaining the tumorigenic ability of cancer cells; inhibitors of BKCa channels may modulate in vitro tumorigenesis via transmembrane depolarization. It is therefore plausible for BKCa channels to be considered putative targets for anticancer therapies.

6 Emilin2

Emilin2 is an extracellular matrix (ECM) protein that exerts antagonistic effects in the tumor microenvironment. By activating the extrinsic apoptotic pathway, Emilin2 affects tumor cell viability [31,32]. It is directly up-regulated by miR-320 and is part of a fibroblast secretome profile that correlates with clinical outcome in breast cancer patients [33,34]. The molecular regulations governed by Emilin2 in breast cancer have been investigated. Marastoni et al. identified Emilin2 as a novel molecular partner of Wnt1 and demonstrated that this interaction led to a significant inhibition of the Wnt signaling pathway. Emilin2 can halt the expression of β-catenin target genes through decreasing LRP6 phosphorylation and β-catenin activation. They also observed that Emilin2 binds to Wnt1 and impairs Wnt signaling activation in vitro and in vivo experiment. Therefore, Emilin2 can slow cell cycle

progression and reduce cell motility, impairing breast cancer cell growth and development [35]. These findings reveal a further mechanism that Emilin2 suppresses tumor growth, providing evidence of the key role of the microenvironment during tumor development and reinforcing the therapeutic potential of this molecule.

7 WISP

WISP1 is located on chromosome 8q24.1–q24.3, contains 5 exons and 4 introns, and is a secreted matrix cellular protein found in the extracellular matrix (ECM) [36]. Human WISP1 was first identified in a human mammary epithelial cell line with Wnt-1 expression and shown to be a Wnt-1-induced gene in 1998 [37]. As well as other ECM proteins, WISP1 affects cell responses, including differentiation, proliferation, migration, and survival [36]. In stromal cells in the proximity of tumors WISP1 overexpression leads to an increase in tumor growth through paracrine signaling [38,39]. Additionally, transfecting WISP1 into melanoma cells inhibited tumor cell growth [40]. Overexpression of WISP1 down-regulated the invasion and migration of lung cancer cells, leading to reduced metastatic potential [41]. However, the investigators found that WISP1 expression were increased in tumor cells in vivo, including colon, lung, liver, and breast cancer [37,42-44]. Chiang et al. discovered that WISP1 functions as an oncogene for human breast cancer. Ectopic expression of WISP1 in breast cancer cells promotes cell growth and metastasis, represses p21 and p27 expression, and stimulates EMT. WISP1, NDRG1, a tumor suppressor gene for breast cancer, is repressed by WISP1 through DNA sequences within the NDRG1 promoter [45]. Thus, WISP1 is a human breast cancer oncogene and is a potential therapeutic target. The epithelial-mesenchymal transition (EMT) has been associated with the acquisition of motility, invasiveness, and self-renewal traits. During both normal development and tumor pathogenesis, this change in cell phenotype is induced by contextual signals that epithelial cells receive from their microenvironment. p21 is a potent cyclin-dependent kinase inhibitor (CKI). The p21 (CIP1/WAF1) protein binds to and inhibits the activity of cyclin-CDK2, -CDK1, and -CDK4/6 complexes, and thus functions as a regulator of cell cycle progression at G1 and S phase. p27 is a cell-cycle regulatory protein that Interacts with cyclin-CDK2 and -CDK4, inhibiting cell cycle progression at G1.

WISP2, a 29-kDa protein, belonging to the cysteine-rich 61/connective tissue growth factor/ nephroblastoma overexpressed (CCN) family [46], was believed to act as a potential proliferation module [47]. The investigators have concluded that WISP2 plays a dual role in the progression of breast and pancreatic cancer, acting as an oncogenic promoter at early stages of tumor development and subsequently, at later stages, as a suppressor of the invasive phenotype [48–50]. Corresponding studies have suggested that less aggressive breast cancer cell lines highly express WISP2, compared to low levels of WISP2 that non-transformed cells express [50]. WISP2 knock-down in less aggressive breast cancer cell lines is accompanied by estrogen-independent cell growth, and isassociated with the loss of estrogen receptor alpha (ERa) expression and increased expression of key components of TGF-β signaling pathway thereby promoting EMT [51], which is similar to WISP1. Furthermore, the researchers suggest that WISP2 can block expression of miR-10b [52], a non-coding RNA known as a role in invasion and metastasis [53]. altogether, these findings suggest that the loss of WISP2 is linked to breast cancer progression [54], accompanied by both EMT induction and increased stemness. These data suggest that WISP2 is a novel target for the development of more efficient therapies toward breast cancer.

8 MicroRNA

MicroRNAs (miRNAs) are small noncoding RNAs that regulate gene expression at a post-transcriptional level and monitor several biological processes [55]. Several human miRNAs have been shown to regulate the metastasis of breast cancer cells [56].

MicroRNA-100 (miR-100) is a member of the miR-100 family of miRNAs and is widely expressed in vertebrates [57]. However, the role of miR-100 in cancers seems to be confounded, since it can act either as an oncogene or as a tumor suppressor in different tumor types [58,59]. In recent years, the investigators discovered that miR-100 functions to suppress breast cancer cell movement and invasion by inhibiting proliferation and survival-promoting oncogene insulin-like growth factor (IGF) 2. In addition, miR-100 can inhibit breast tumorigenesis [60] and target HOXA1 [61]. HoxA1 is transcriptionally regulated by retinoic acid (RA) and encodes a transcription factor which has been shown to play important roles in cell differentiation and embryogenesis. Jiang et al observed that the overexpression of MiR-100 could inhibit the migration and invasion of breast cancer cells by transfecting miR-100 mimic in aggressive breast cancer cell lines and transfecting miR-100 inhibitor in non-metastatic cell lines. This mechanism involves MiR-100 directly inhibiting the expression of FZD-8 and inactivating the

Wnt/β-catenin pathway in breast cancer cells. MiR-100 functions as a tumor suppressor in breast cancer cells and the manipulation of miR-100 provides a promising therapeutic strategy for breast cancer treatment.

MiR-340 has been studied as a putative tumor suppressor in several cancers including neurofibromatosis type 1, neuroblastoma, ovarian tumor, and gastric cancer [62-64]. The investigators have shown that MiR-340 suppresses cell migration, invasion, and metastasis in these cancers due to its over-expression [65]. Moreover, in vitro, the investigators confirmed that over-expression of MiR-340 regulates motility of cancer cells and decreases cell mobility and invasion. The restoration of miR-340 expression presents a novel therapeutic strategy for preventing breast cancer progression and metastasis if we undertake more comprehensive investigations and trials.

MiRNA-301a has attracted much attention due to its important role in various biological and pathological processes, including development, differentiation, inflammation, apoptosis and cancer [66-68]. Ma et al. have shown that MiR-301a is involved with breast cancer development and metastasis by directly targeting PTEN to activate the Wnt/β-catenin pathway, revealing the oncogenic role of miR-301a in breast cancer. Inhibition of miR-301 presents a promising therapeutic strategy for breast cancer treatment

9 NRBP1

NRBP1 is a ubiquitously expressed adapter protein [69]. Recently it has been discovered that it can suppress tumors [70] in cytoplasm and nucleus, where it has been detected. Additionally, it has been demonstrated to predominantly localize in the cytoplasm. In vitro studies have suggested that NRBP1 shuttles between the nucleus and cytoplasm, functioning to regulate protein localization and undertake transcription factor activity. NRBP1 has also been implicated with cancer development. Not only were NRBP1 levels reduced in breast cancer tumor tissues, but NRBP1 expression level and breast cancer clinic pathological features were correlated in patients. Importantly, the Wnt signaling pathway could regulate NRBP1-induced cancer cell proliferation. Based on this information, NRBP1 could be a potential therapeutic target for suppressing breast cancer metastasis.

10 TRAF4

Tumor necrosis factor receptor-associated factor 4 (TRAF4) is a member of the TRAF family, whose members act as adaptors in several receptor-mediated signaling pathways [71]. Reports have indicated that TRAF4 can enhance transcription of β-catenin and may protect it from p53-mediated degradation [72]. A review indicated that TRAF4 had a negligible effect on Wnt in early *Xenopus* embryonic tissue [8]. Studies have shown that the TRAF4 is highly expressed in breast cancer tissue. In a similar manner as β-catenin it can promote cell migration and metastasis in breast cancer [73,74]. Wang et al. discovered that TRAF4 can bind to β-catenin and enhance expression of β-catenin; in addition, they also found that TRAF4 mediated the translocation of β-catenin from the cytoplasm to the nucleus, thereby facilitating activation of the Wnt signaling pathway in breast cancer.

11 Wntless (Wls)/Evi/Sprinter/GPR177

Wntless (Wls)/Evi/Sprinter/GPR177 is a seven-pass transmembrane protein, which is highly conserved and localized to compartments of the secretory pathway among vertebrates including the Golgi apparatus, endosomes, and plasma membrane [75,76]. As a Wnt cargo receptor, Wls shuttles palmitoylated Wnts from the endoplasmic reticulum to the plasma membrane, and is also required for exocytosis of Wnt proteins from the Wnt-producing cells [77-79]. Knockdown of Wls leads to an accumulation of Wnts in the producing cells [80], resulting in early embryonic patterning defects [81]. Moreover, Wls deficiency impairs mammary development which interferes with mammary stem cells, causing deficiencies in cell proliferation and differentiation [82]. We found that Wls has been overexpressed in several kinds of cancers, such as glioblastoma, colorectal cancer, B cell precursor acute lymphoblastic leukemia (BCP ALL), ovarian cancer, and gastric cancer in the past few years [83-85]. Additionally, Wls could promote glioma cell proliferation and invasion through regulating Wnt secret ion and upregulation of interleukins and other pro-oncogenic factors [82]. Some studies demonstrate that Wls inhibits melanoma cell proliferation through the β-catenin signaling pathway [86]. The strong Wls expression observed in cancer suggests a potential role for Wls in breast tumorigenesis. Downregulation of Wls could reduce colony formation and tumor cell growth through inhibiting the secretion of Wnt and its downstream signaling. Our results indicate that Wls might be able to promote proliferation of breast cancer cells and may provide a new therapeutic target for breast cancer.

12 Conclusion

The Wnt signaling pathway plays an integral role in malignant cell growth, proliferation, motility, and survival of tumors, and is widely observed in breast cancer. As previously summarized, an increasing number of new targets have been identified that inhibit Wnt signaling. Much work remains to be done in to apply this new information to clinical treatments and to develop novel Wnt signaling inhibitors. XAV-939 can selectively inhibit the transcription mediated by Wnt/β-catenin through inhibiting tankyrase1/2. ICG-001 suppress the transcription mediated by Wnt/β-catenin/TCF and selectively interacting promoter binding protein. Overall, the diversity and rationale behind the use of Wnt/β-catenin targets support the Wnt signaling inhibitors as promising therapeutics. Although most drugs are still at a very early developmental stage, the importance of this pathway makes breast cancer a strong candidate to benefit from these new therapies. It is important not to oversell the promise of Wnt signaling-based therapies, but continued research will, we believe, help to solve some of these vexing issues. The result is certain to be exciting, and will lead to new insights that translate to better therapies for breast cancer.

Conflict of interest: Authors declare nothing to disclose.

References

[1] Jemal A., Bray F., Melissa M., Ferlay J., Ward E., Forman D., et al., Global cancer statistics, Can. Cancer J. Clin., 2011, 61, 69–90.

[2] Ferlay J., Steliarova-Foucher E., Lortet-Tieulent J., Rosso S., Coebergh J.W.W., Comber H., et al., Cancer incidence and mortality patterns in Europe: estimates for 40 countries in 2012, Eur. J. Cancer, 2013, 49, 1374–403.

[3] Cai J., Guan H., Fang L., Yang Y., Zhu X., Yuan J., et al., MicroRNA-374a activates Wnt/β-catenin signaling to promote breast cancer metastasis, J. Clin. Invest., 2013, 123, 566–579.

[4] Tian X., Liu Z,, Niu B., Zhang J., Tan T., Lee S., et al., E-cadherin/β-catenin complex and the epithelial barrier, J. Biomed. Biotechnol., 2011, 567305.

[5] MacDonald B.T., Tamai K., He X, Wnt/beta-catenin signaling: components, mechanisms, and diseases, Dev. Cell, 2009, 17, 9–26.

[6] Struewing I., Boyechko T., Barnett C., Beildeck M., Byers S.W., Mao C.D., et al, The balance of TCF7L2 variants with differential activities in Wnt-signaling is regulated by lithium in a GSK3 beta-independent manner, Biochem. Biophys. Res. Commun., 2010, 399, 245–250.

[7] Thompson J., Wong L., Lau P.S., Bannigan J, Adherens junction breakdown in the periderm following cadmium administration in the chick embryo: distribution of cadherins and associated molecules, Reprod. Toxicol., 2008, 25,39–46.

[8] Baarsma H.A., Konigshoff M., Gosens R, The WNT signaling pathway from ligand secretion to gene transcription: molecular mechanisms and pharmacological targets, Pharmacol. Ther., 2013, 138, 66–83.

[9] Caliceti C., Nigro P., Rizzo P., Ferrari R, ROS, Notch, and Wnt signaling pathways: crosstalk between three major regulators of cardiovascular biology, Biomed. Res. Int., 2014, 2014, 318714.

[10] Bin-Nun N., Lichtig H., Malyarova A., Levy M, Elias S, Frank D., et al, PTK7 modulates Wnt signaling activity via LRP6, Development, 2014, 141, 410–412.

[11] Rahmani M., Carthy J.M., McManus B.M, Mapping of the Wnt/beta-catenin/TCF response elements in the human versican promoter, Methods. Mol. Biol., 2012, 836, 35–52.

[12] White B.D, Chien A.J, Dawson D.W Dysregulation of Wnt/beta-catenin signaling in gastrointestinal cancers, Gastroenterology, 2012, 142, 219–232.

[13] De A, Wnt/Ca2+ signaling pathway: a brief overview, Acta. Biochim. Biophys. Sin., 2011, 43, 745–56.

[14] Dawson K., Aflaki M., Nattel S, Role of the Wnt-Frizzled system in cardiac pathophysiology: a rapidly developing, poorly understood area with enormous potential, J. Physiol., 2013, 591, 1409–1432.

[15] Nagase T., Shimozawa N., Takemoto Y., Suzukib Y., Komoric M., Kondoa N., et al, Peroxisomal localization in the developing mouse cerebellum: implications for neuronal abnormalities related to deficiencies in peroxisomes, Biochim. Biophys. Acta., 2004, 1671,26–33.

[16]Zaslavsky A., Chou S.T., Schadler K., Lieberman A., Pimkin M., Kim Y., et al, The calcineurin-NFAT pathway negatively regulates megakaryopoiesis, Blood, 2013, 121, 3205–3215.

[17] Liu P.C., Thiele D.J, Novel stress-responsive genes EMG1 and NOP14 encode conserved, interacting proteins required for 40S ribosome biogenesis, MOL. BIOL. CELL, 2001, 12, 3644–3657.

[18] Munoz I.M., Jowsey P.A., Toth R., Rouse J, Phospho-epitope binding by the BRCT domains of hPTIP controls multiple aspects of the cellular response to DNA damage, NUCLEIC. ACIDS. RES., 2007, 35, 5312–5322.

[19] Gong Z., Cho Y.W., Kim J.E., Ge K., Chen J.J, Accumulation of Pax2 transactivation domain interaction protein (PTIP) at sites of DNA breaks via RNF8-dependent pathway is required for cell survival after DNA damage, J. BIOL. CHEM., 2009, 284, 7284–7293.

[20] Cho E.A., Prindle M.J., Dressler G.R., BRCT domain containing protein PTIP is essential for progression through mitosis, Mol. Cell. Biol., 2003, 23, 1666–1673.

[21] Woods N.T., Mesquita R.D., Sweet M., Marcelo A., Carvalho, Li X.L., et al, Charting the landscape of tandem BRCT domain-mediated protein interactions. Sci. Signal, 2012, 5, s6.

[22] Havugimana P.C., Hart G.T., Nepusz T., Yang H., Turinsky A.L.,Li Z., et al, A census of human soluble protein complexes, CELL, 2012, 150, 1068–1081.

[23] Cao Q., Mani R.S., Ateeq B., Dhanasekaran S.M., Asangani I.A., Prensner J.R., et al, Coordinated regulation of polycomb group complexes through microRNAs in cancer, Cancer Cell, 2011, 20, 187–199.

[24] Cao Q., Wang X., Zhao M., Yang R., Malik R., Qiao Y., et al, The central role of EED in the orchestration of polycomb group complexes, Nat. Commun., 2014, 5, 3127.
[25] Lei J.J., Peng R.J., Kuang B.H., Yuan Z.Y., Qin T., Liu W.S., et al, NOP14 suppresses breast cancer progression by inhibiting NRIP1/Wnt/β-catenin pathway, Oncotarget, 2015, 6, 25701-25714.
[26] Khaitan D., Sankpal U.T., Weksler B., Meister E.A., Romero I.A., Couraud P.O., et al, Role of KCNMA1 gene in breast cancer invasion and metastasis to brain, BMC Cancer, 2009, 9, 258.
[27] Liu X., Chang Y., Reinhart P.H., Sontheimer H, Cloning and characterization of glioma BK, a novel BK channel isoform highly expressed in human glioma cells, J. Neurosci., 2002, 22, 1840-1849.
[28] Lu R., Alioua A., Kumar Y., Toro L, MaxiK channel partners: physiological impact, J. Physiol., 2006, 570, 65-72.
[29] Tian L., McClafferty H., Chen L., Shipston M.J, Reversible tyrosine protein phosphorylation regulates large conductance voltage- and calcium-activated potassium channels via cortactin, J. Biol. Chem., 2008, 283, 3067-3076.
[30] So E.C., Wu K.C., Liang C.H., Chen J.Y., Wu S.N, Evidence for activation of BKCa channels by a known inhibitor of focal adhesion kinase, PF573228, Life Sci., 2011, 89, 691-701.
[31] Mongiat M., Ligresti G., Marastoni S., Doliana R., Colombatti A, Regulation of the extrinsic apoptotic pathway by the extracellular matrix glycoprotein EMILIN2, Mol. Cell. Biol., 2007, 27, 7176–7187.
[32] Mongiat M., Marastoni S., Ligresti G., Lorenzon E., Schiappacassi M., Perris R, The extracellular matrix glycoprotein elastin microfibril interface located protein 2: a dual role in the tumor microenvironment, Neoplasia, 2010, 12, 294–304.
[33] Hill V.K., Hesson L.B., Dansranjavin T., Dallol A., Bieche I., Vacher S., et al, Identification of five novel genes methylated in breast and other epithelial cancers, Mol. Cancer, 2010, 9, 51.
[34] Bronisz A., Godlewski J., Wallace J.A., Merchant A., Nowicki M., Mathsyaraja H., et al, Reprogramming of the tumor microenvironment by stromal PTEN-regulated miR-320, Nat. Cell Biol., 2012, 14, 159-167.
[35] Marastoni S., Andreuzzi E., Paulitti A., Colladel R., Pellicani R., Todaro F., et al, EMILIN2 down-modulates the Wnt signalling pathway and suppresses breast cancer cell growth and migration, J. Pathol., 2014, 232, 391-404.
[36] Chen C.C., Lau L.F., Functions and mechanisms of action of CCN matricellular proteins, Int. J. Biochem. Cel. Biol., 2009, 41, 771-783.
[37] Pennica D., Swanson T.A., Welsh J.W., Roy M.A., Lee L.J., et al, WISP genes are members of the connective tissue growth factor family that are up-regulated in wnt-1-transformed cells and aberrantly expressed in human colon tumors, Proc. Natl. Acad. Sci. USA., 1998, 95, 14717–14722.
[38] Hsu J.Y., Reimann J.D., Sorensen C.S., Lukas J., Jackson P.K., E2F-dependent accumulation of hEmi1 regulates S phase entry by inhibiting APC (Cdh1), Nat. Cell Biol., 2002, 4, 358–366.
[39] Tanaka S., Sugimachi K., Saeki H., Kinoshita J., Ohga T., Shimada M., et al, A novel variant of WISP1 lacking a Von Willebrand type C module overexpressed in scirrhous gastric carcinoma, Oncogene, 2001, 20, 5525–5532.
[40] Hashimoto Y., Okada N.S., Tani M., Nagamachi Y., Takeuchi K., Shiroishi T., et al,Expression of the Elm1 gene, a novel gene of the CCN (connective tissue growth factor, Cyr61/Cef10, and neuroblastoma overexpressed gene) family, suppresses In vivo tumor growth and metastasis of K-1735 murine melanoma cells, J. Exp. Med., 1998, 187, 289–296.
[41] Soon L., Yie T.A., Shvarts A., Levine A.J., Su F., Tchou-Wong K.M., et al, Overexpression of WISP-1 down-regulated motility and invasion of lung cancer cells through inhibition of Rac activation, J. Biol. Chem., 2003, 278, 11465–11470.
[42] Calvisi D.F., Conner E.A., Ladu S., Lemmer E.R., Factor V.M., Thorgeirsson S.S., et al, Activation of the canonical Wnt/beta-catenin pathway confers growth advantages in c-Myc/E2F1 transgenic mouse model of liver cancer, J. Hepatol., 2005, 42, 842–849.
[43] Margalit O., Eisenbach L., Amariglio N., Kaminski N., Harmelin A., Pfeffer R., et al, Overexpression of a set of genes, including WISP-1, common to pulmonary metastases of both mouse D122 Lewis lung carcinoma and B16-F10.9 melanoma cell lines, Brit. J. Cancer, 2003, 89, 314–319.
[44] Xie D., Nakachi K., Wang H., Elashoff R., Koeffler H.P., et al, Elevated levels of connective tissue growth factor, WISP-1, and CYR61 in primary breast cancers associated with more advanced features, Cancer Res., 2001, 61, 8917–8923.
[45] Chiang K.C., Yeh C.N., Chung L.C., Feng T.H., Sun C.C., Chen M.F., et al, WNT-1 inducible signaling pathway protein-1 enhances growth and tumorigenesis in human breast cancer, SCIENTIFIC REPORTS, 2015, 5, 1-11.
[46] Brigstock D.R, The CCN family: a new stimulus package J. Endocrinol., 2003, 178, 169–175.
[47] Leask A., Abraham D.J, All in the CCN family: essential matricellular signaling modulators emerge from the bunker, J. Cell. Sci., 2006, 119, 4803–4810.
[48] Banerjee S., Dhar G., Haque I., Kambhampati S., Mehta S., Sengupta K., et al, CCN5/WISP-2 expression in breast adenocarcinoma is associated with less frequent progression of the disease and suppresses the invasive phenotypes of tumor cells, Cancer. Res., 2008, 68, 7606–7612.
[49] Dhar G., Mehta S., Banerjee S., Banerjee S.K, Loss of WISP-2/CCN5 signaling in human pancreatic cancer: a potential mechanism for epithelial-mesenchymal-transition, Cancer Lett., 2007, 254, 63–70.
[50] Fritah A., Saucier C., De Wever O., Bracke M., Bièche I., Lidereau R., et al, Role of WISP-2/CCN5 in the maintenance of a differentiated and noninvasive phenotype in human breast cancer cells, Mol. Cell. Biol., 2008, 28, 1114–1123.
[51] Sabbah M., Prunier C., Ferrand N., Megalophonos V., Lambein K., De Wever O., et al, CCN5, a novel transcriptional repressor of the transforming growth factor beta signaling pathway, Mol. Cell Biol., 2011, 31, 1459–1469.
[52]Haque I., Banerjee S., Mehta S., De A., Majumder M., Mayoet M.S., et al, Cysteine-rich 61-connective tissue growth factor-nephroblastoma-overexpressed 5 (CCN5)/ Wnt-1-induced signaling protein-2 (WISP-2) regulates microRNA-10b via hypoxia-inducible factor-1alpha-TWIST signaling networks in human breast cancer cells, J. Biol. Chem., 2011, 286, 43475–43485.
[53] Ma L., Teruya-Feldstein J., Weinberg R.A., Tumor invasion and metastasis initiated by microRNA-10b in breast cancer, Nature, 2007, 449, 682–688.
[54] Banerjee S.K., Banerjee S., CCN5/WISP-2: A micromanager of breast cancer progression, J. Cell. Commun. Signal., 2012, 6, 63–71.

[55] So W.O., Hwang D.W., Lee D.S., In vivo Monitoring of microRNA Biogenesis Using Reporter Gene Imaging, Theranostics, 2013, 3, 1004–1011.
[56] Vimalraj S., Miranda P.J., Ramyakrishna B, Regulation of Breast Cancer and Bone Metastasis by MicroRNAs, Dis. Markers, 2013, 35, 369–387.
[57] Christodoulou F., Raible F., Tomer R., Arendt D., Ancient animal microRNAs and the evolution of tissue identity, Nature, 2010, 463, 1084–1088.
[58] Zheng Y.S., Zhang H., Zhang X.J., Feng D.D.; Luo X.Q. Zeng C.W., et al, MiR-100 regulates cell differentiation and survival by targeting RBSP3, a phosphatase-like tumor suppressor in acute myeloid leukemia, Oncogene, 2012, 31, 80–92.
[59] Li Z.P., Li X., Yu C., Wang M., Peng F., Xiao J., et al, MicroRNA-100 regulates pancreatic cancer cells growth and sensitivity to chemotherapy through targeting FGFR3, Tumor. Biol., 2014, 35, 11751–11759.
[60] Gebeshuber C.A., Martinez J., miR-100 suppresses IGF2 and inhibits breast tumorigenesis by interfering with proliferation and survival signaling, Oncogene, 2013, 32, 3306–3310.
[61] Chen D., Sun Y., Yuan Y., Han Z., Zhang P., Zhang J., et al, miR-100 induces epithelial-mesenchymal transition but suppresses tumorigenesis, migration and invasion, PLoS Genet., 2014, 10, e1004177.
[62] De C.L., Berardi M., Sommariva M., Cataldo A., Canevari S., Mezzanzanica D., et al, Increased sensitivity to chemotherapy induced by CpG-ODN treatment is mediated by microRNA modulation, PLoS One, 2013, 8, e58849.
[63] Presneau N., Eskandarpour M., Shemais T., Henderson S., Halai D., Tirabosco R., et al, MicroRNA profiling of peripheral nerve sheath tumors identifies miR-29c as a tumor suppressor gene involved in tumor progression, Br. J. Cancer, 2013, 108, 964–972.
[64] Das S., Bryan K., Buckley P.G., Piskareva O., Bray I.M., Foley N., et al, Modulation of neuroblastoma disease pathogenesis by an extensive network of epigenetically regulated microRNAs, Oncogene, 2013, 32, 2927–2936.
[65] Wu Z.S., Wu Q., Wang C.Q., Wang X.N., Huang J.; Zhao J.J., et al, miR-340 inhibition of breast cancer cell migration and invasion through targeting of oncoprotein c-Met, Cancer, 2011, 117, 2842-2852.
[66] Lee W.A., Na Y.S., Jeong S.y., Jeong S.R., Park H.R. , Chuang J., et al, Comparison of inflammatory microRNA expression in healthy and periodontitis tissues, Biocell, 2011, 35, 43–49.
[67] Wang Z, The concept of multiple-target anti-miRNA antisense oligonucleotide technology, Methods Mol. Biol., 2011, 676: 51-57.
[68] Xu X.D., He X.J.; Tao H.Q.; Zhang W.; Wang Y.Y.; Ye Z.Y., et al, Abnormal expression of miR-301a in gastric cancer associated with progression and poor prognosis, J. Surg. Oncol., 2013, 108,197–202.
[69] Wilson C.H., Crombie C., van der Weyden L., George P., Alistair G.R., Mercedes P., et al, Nuclear receptor binding protein 1 regulates intestinal progenitor cell homeostasis and tumor formation, EMBO J., 2012, 31, 2486–2497.
[70] Kerr J.S., Wilson C.H., Nuclear receptor-binding protein 1: a novel tumour suppressor and pseudokinase, Biochem. Soc. Trans., 2013, 41, 1055–1060.
[71] Kedinger V., Rio M.C., TRAF4, the unique family member, Adv. Exp. Med. Biol., 2007; 597:60–71.
[72] Rozan L.M., El-Deiry W.S., Identification and characterization of proteins interacting with Traf4, an enigmatic p53 target, Cancer Biol. Ther., 2006, 5, 1228–1235.
[73] Zhang L., Zhou F., Vinuesa G.D., Esther M., Mesker W.E., Li H., et al, TRAF4 promotes TGF-b receptor signaling and drives breast cancer metastasis, Mol. Cell., 2013, 51, 559–572.
[74] Wang X., Jin C., Tang Y., Tang L.Y., Zhang Y.E., Ubiquitination of tumor necrosis factor receptor associated factor 4 (TRAF4) by smad ubiquitination regulatory factor 1 (Smurf1) regulates motility of breast epithelial and cancer cells, J. Biol. Chem., 2013, 288, 21784–21792.
[75] Banziger C., Soldini D., Schutt C., Peder Z., George H., Konrad B., Wntless, a conserved membrane protein dedicated to the secretion of Wnt proteins from signaling cells, Cell, 2006, 125, 509–522.
[76] Yu J., Chia J., Canning C.A., Jones C.M., Bard F.A., Virshup D.M., WLS retrograde transport to the endoplasmic reticulum during Wnt secretion, Dev. Cell., 2014, 29, 277–291.
[77] Eaton S., Retromer retrieves Wntless, Dev. Cell., 2008; 14, 4–6.
[78] Bartscherer K., Pelte N., Ingelfinger D., Michael B., Secretion of Wnt ligands requires Evi, a conserved transmembrane protein, Cell, 2006, 125, 523–533.
[79] Hausmann G., Banziger C., Basler K., Helping wingless take flight: how WNT proteins are secreted, Nat. Rev. Mol. Cell. Biol., 2007, 8, 331–336.
[80] Fu J., Jiang M., Mirando A.J., Ivy Yu H.M., Wei Hsu., Reciprocal regulation of Wnt and Gpr177/mouse Wntless is required for embryonic axis formation, Proc. Natl. Acad. Sci. USA., 2009, 106, 18598–603.
[81] Maruyama E.O., Yu H.M., Jiang M., Jiang Fu., Wei Hsu., Gpr177 deficiency impairs mammary development and prohibits Wnt-induced tumorigenesis, PLoS One, 2013, 8, e56644.
[82] Augustin I., Goidts V., Bongers A., Kerr G., Vollert G., Radlwimmer B., et al, The Wnt secretion protein Evi/Gpr177 promotes glioma tumourigenesis, EMBO. Mol. Med., 2012, 4, 38–51.
[83] Voloshanenko O., Erdmann G., Dubash T.D., Iris A. , Marie M., Giusi M., et al, Wnt secretion is required to maintain high levels of Wnt activity in colon cancer cells, Nat. Commun., 2013, 4, 2610.
[84] Stewart J., James J., McCluggage G.W., Manuel S.T., Analysis of Wntless (WLS) expression in gastric, ovarian, and breast cancers reveals a strong association with HER2 overexpression, Mod. Pathol., 2015, 28,428-436.
[85] Chiou S.S., Wang L.T., Huang S.B., Chai C.Y., Wang S.N., Liao Y.M., et al, Wntless (GPR177) expression correlates with poor prognosis in B-cell precursor acute lymphoblastic leukemia via Wnt signaling, Carcinogenesis, 2014, 35, 2357-2364.
[86] Yang P.T., Anastas J.N., Toroni R.A., Shinohara M.M., Goodson J.M., Bosserhoff A.K., et al, WLS inhibits melanoma cell proliferation through the ss-catenin signaling pathway and induces spontaneous metastasis, EMBO Mol. Med., 2012, 4, 1294-307.