Am J Transl Res 2018;10(7):1949-1960
www.ajtr.org /ISSN:1943-8141/AJTR0079628

Review Article

Gankyrin as a potential target for tumor therapy: evidence and perspectives

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Received January 13, 2018; Accepted May 16, 2018; Epub July 15, 2018; Published July 30, 2018

Abstract: Gankyrin (also known as PSMD10 or p28GANK), engages in diverse biological processes, including cellular growth, proliferation and invasion. Several studies have demonstrated that Gankyrin is a candidate oncogene. In parallel, the dysregulation of Gankyrin has been observered in a variety of human cancer. Overexpression of Gankyrin is involved in tumor initiation and progression by regulating several signaling pathways that control cell-cycle process, cell growth, apoptosis, et al. On the contrary, downregulation of Gankyrin significantly inhibits cell growth, proliferation and metastasis. Therefore, Gankyrin appears to be a potential target for tumor therapy. Herein, this review summarizes the current knowledge in understanding the biological functions and oncogenic role of Gankyrin in human cancers from the perspective of clinical-pathological significances, aiming to provide guidance for the development of Gankyrin-targeted therapy.

Keywords: Gankyrin, target, tumor therapy

Introduction

Gankyrin is localized to human chromosome Xq22.3 and has been highly conserved throughout evolution [1, 2]. Gankyrin is reported to play important roles in cellular growth, proliferation, invasion and metastasis [3]. An increasing number of studies have shown that Gankyrin is prominently overexpressed in the vast majority of human malignancies [4-7]. Moreover, the abnormal expression of Gankyrin has also been proven to be significantly related to the clinicopathological parameters of the disease, highlighting the important function of Gankyrin as a biomarker in different cancers. Herein, this review will summarize the biological functions and oncogenic roles of Gankyrin, particularly focusing on Gankyrin aberrations and the consequences of its deregulation in tumorigenesis and development, with the aim of assessing the potential application of this protein as a drug target for cancer treatment.

Brief overview of biological functions of Gankyrin

Gankyrin was originally isolated by Higashitsuji et al. through cDNA subtractive hybridization in human hepatocellular carcinoma [1, 2]. It was initially identified as a component of the 26S proteasome and consists of 7 ankyrin repeats, which are involved in protein-protein interactions [8-10]. Gankyrin has impacts on diverse biological processes, such as cellular growth, proliferation, and invasion, and it contributes to oval cell-mediated liver regeneration and cell cycle progression [3]. Overexpression of Gankyrin in NIH-3T3 cells leads to the transformation of those cells and inoculation of those transformed cells into nude mice results in tumor formation [11]. The role of Gankyrin as a tumor oncogene has been established in different types of human cancers, and Gankyrin could regulate tumor suppressors post-translationally, degrading the proteins. By interaction with retinoblastoma protein (Rb) and cyclin-dependent kinase 4 (CDK4), Gankyrin promotes Rb phosphorylation and inactivation, and activates the E2F transcription factor, leading to cell cycle progression [12-14]. Additionally, Gankyrin increases the ubiquitylation and degradation of p53 by binding to murine double minute 2 (Mdm2) [15]. Furthermore, Gankyrin plays a critical role in Ras-initiated carcinogenesis and regulates Ras-mediated activation of AKT through the inhibition of the RhoA/ROCK/PTEN

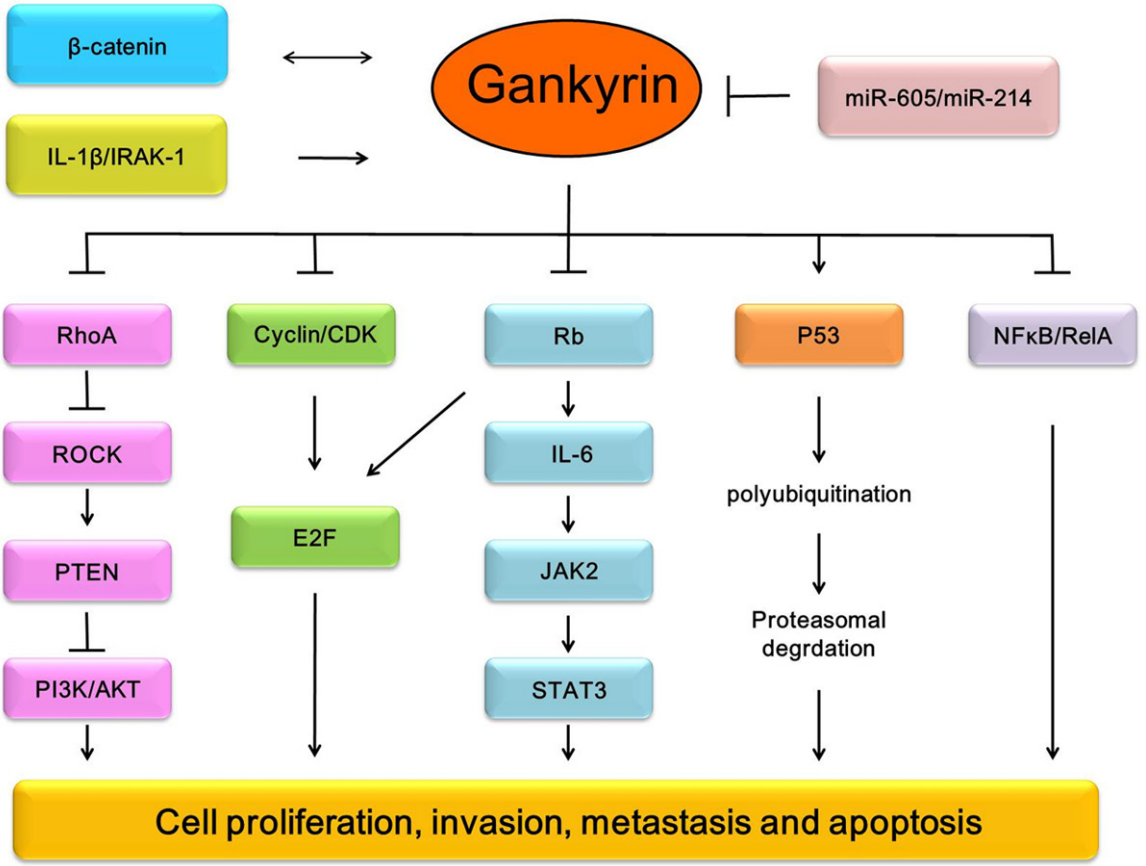

Figure 1. Several Proposed important schematic signaling pathways for Gankyrin in tumorigenesis. Abnormal expression of Gankyrin affects tumorigenesis process, such as cell proliferation, invasion, metastasis and apoptosis by RhoA/Rock/PTEN/PI3K/AKT pathway or other signaling cascades. Abbreviation: IL-1β: Interleukin 1 beta; IRAK-1: Interleukin-1 receptor associated kinase 1; RhoA: Ras homolog gene family, member A; ROCK: Rho-associated protein kinase; PTEN: Phosphatase and tensin homolog; CDK: Cyclin-dependent kinase; IL-6: Interleukin 6; JAK2: Janus kinase 2; STAT3: signal transducer and activator of transcription 3; NFκB: nuclear factor kappa light chain enhancer of activated B cells; RelA: Transcription factor p65, also known as nuclear factor NF-kappa-B p65 subunit.

signaling pathway [16]. Recent reports have demonstrated that Gankyrin mediates cell proliferation through the activation of the PI3K/AKT or IL-6/STAT3 pathways [6, 17]. Moreover, it is reported that Gankyrin forms a positive feedback loop in β-catenin signaling. Gankyrin is regulated by β-catenin signaling and the overexpression of Gankyrin positively enhances β-catenin transcription activity by reducing E-cadherin protein levels and membrane localization of the E-cadherin/β-catenin complex, indicating that β-catenin may be a critical transcriptional activator of Gankyrin [18]. Gankyrin may inhibit NFκB/RelA at the transcriptional level by binding directly to RelA and modulating RelA acetylation via SIRT1 [19]. On the other hand, another report demonstrated that Gankyrin inhibited the nuclear translocation of NFκB/RelA and suppressed its activity [10]. It has also been reported that Gankyrin interacts with the IL-1β/IRAK-1 inflammatory signaling pathway. Activation of the IL-1β/IRAK-1 pathway induces the binding of the nuclear factor (NF-Y) complex to the Gankyrin promoter, facilitates the recruitment of the coactivators E1A-binding protein p300 (p300) and CREB-binding protein (CBP), and finally increases Gankyrin expression [20]. In addition, Gankyrin could be regulated by miRNA at the posttranscriptional level; a recent report found that miR-605 could directly bind to the 3'UTR of Gankyrin, significantly reducing its expression in intrahepatic cholangiocarcinoma (ICC), and the overexpression of miR-605 repressed ICC cell proliferation and invasion by downregulating Gankyrin. Moreover, the restoration of Gankyrin protein expression counteracted the cell phenotypic alterations caused by miR-605 [21]. Another

Table 1. Functions of Gankyrin in different human cancers

Cancer	Expression	Phenotypes of affected	Related genes/pathways	References
Hepatocellular cancer	Upregulated	Cell cycle, proliferation, metastasis, stem cells self-renewal capacity, autophagy	STAT3/AKT, HNF4α, ATG7, Nrf2	[4, 5, 25-31]
Glioma	Upregulated	Proliferation	Uncertain	[32]
Oral Cancer	Upregulated	Proliferation	Uncertain	[33]
Lung cancer	Upregulated	Proliferation, metastasis, EMT	PI3K/AKT, IL-6/STAT3, TGFβ/SMAD3	[16, 34, 35]
Esophageal cancer	Upregulated	Cell growth, motility, invasiveness, metastasis	Uncertain	[36, 37]
Gastric cancer	Upregulated	Cell growth, proliferation, chemoresistence	cyclin D1, cyclin E, PI3K/AKT	[38-41]
Colorectal cancer	Upregulated	Proliferation, metastasis	IL-8, cyclin D1, mTORC1, PI3K/GSK-3β/β-catenin, TNF-α, IL-7, STAT3	[42-47]
Pancreatic cancer	Upregulated	Cell growth, proliferation, cell cycle	cyclin A, cyclin D1, cyclin E, CDK2, CDK4, PCNA, Rb, P27, P53	[48]
Liposarcoma	Upregulated	Cell proliferation, colony-formation, migration and tumorigenesis	P53, P21, AKT/mTOR	[50]
Cervical cancer	Upregulated	Tumorigenesis, metastasis, EMT	Vimentin, β-catenin and Twist2, E-cadherin	[51]
Ovarian cancer	Upregulated	Proliferation	PI3K/AKT, cyclin D1	[53]
Endometrial cancer	Upregulated	Proliferation	PTEN/PI3K/AKT	[6]
Breast cancer	Upregulated	Tumorigenesis, invasiveness, metastasis	Rac1, E-cadherin	[7, 54, 55]
Cholangiocarcinoma	Upregulated	Cell growth, proliferation, invasion, metastasis	IL-6/STAT3, miR-605	[17, 21]
Testicular germ cell tumors	Upregulated	Cell proliferation	Rb	[57]
Prostate cancer	Upregulated	Tumorigenesis	Uncertain	[58]

study has shown that ectopic expression of miR-214 inhibits myeloma cell growth and induces apoptosis by the downregulation of the Gankyrin protein [22]. Therefore, repressing Gankyrin activity has been identified as a potential anticancer strategy. **Figure 1** presents important oncogenic mechanisms of Gankyrin.

Gankyrin functions as an oncogene in hepatocellular carcinomas and a variety of other cancers

Here, we review the evidence supporting Gankyrin as an oncogene in hepatocellular carcinoma and many other cancers, including gliomas, liposarcoma and cancers of the lung, esophagus, stomach, colon, pancreas, cervix, ovary, endometrium, and breast (**Table 1**). Emerging studies have revealed that intensive expression of Gankyrin may be crucial for the achievement of invasive and/or aggressive phenotypes in various carcinomas, and a growing number of studies have concluded that Gankyrin may be a potential biomarker for cancer diagnosis and a therapeutic target for cancer treatment.

Role of Gankyrin in hepatocellular carcinomas

Hepatocellular cancer (HCC) is an aggressive malignancy and has an increasing incidence worldwide [23]. The outcome for advanced stage HCC patients remains extremely poor due to the high rates of relapse and metastasis. Conventional therapies, including hepatectomy, transplantation, and chemotherapy, have shown limited efficacy in HCC patients with advanced disease [24]. The heterogeneity among individuals and tumors may contribute to treatment failure; precise treatment is thus required, and the underlying molecular events that are responsible for HCC initiation and progression provide us with more effective targeted therapies for individualized treatment.

Gankyrin levels have been shown to be gradually elevated throughout the consecutive stages of hepatocarcinogenesis [4, 5]. Fu et al. reported that Gankyrin expression was absent in normal liver tissue, was weakly expressed in liver cirrhosis and paracarcinoma liver tissues, and was strongly expressed in HCC tissue. Additionally, Gankyrin expression was significantly correlated with portal vein tumor thrombus and microscopic hepatic vein involvement, indicating that Gankyrin might contribute to HCC onset, progression and metastasis [4]. In line with these findings, Jing et al. also found that the expression level of Gankyrin was significantly higher in hepatocellular cancer than in normal and benign tissues, and upregulated Gankyrin expression was significantly associated with capsular invasion and intrahepatic metastasis; however, there was no correlation between Gankyrin expression and tumor size, differentiation status, the number of tumors, intravascular tumor thrombus, capsule formation, lymph node metastases, or overall survival. Furthermore, silencing Gankyrin in the HepG2 cell line inhibited tumorigenesis and metastasis. These findings indicated that Gankyrin could be a useful diagnostic marker for HCC and could play an important role in tumor initiation, promotion and progression [5]. Surprisingly, contrary to Jing's findings, another study found that Gankyrin expression was significantly higher in patients with increased tumor size, vascular invasion, and intrahepatic and distant metastasis, and high Gankyrin levels were correlated with unfavorable outcomes [25]. Thus, further comprehensive investigations will be necessary to validate the oncogenic events related to Gankyrin in hepatic cancer. An *in vivo* study of HCC mouse models revealed that Gankyrin ablation in nonparenchymal cells repressed the activity of STAT3, production of IL-6 and expression levels of cancer stem cell markers (such as Bmi1 and epithelial cell adhesion molecule [EpCAM]), reducing tumorigenic capacity [26]. What's more, the study found that the expression of Gankyrin in the tumor microenvironment was inversely related with progression free survival (PFS) in patients with HCC who received sorafenib treatment; identification of responders to sorafenib by analyzing Gankyrin expression may help to increase the benefits of this treatment [26].

The Gankyrin/STAT3/AKT pathway can be inhibited by LBH489 (panobinostat), which is a novel hydroxamic acid-derived histone deacetylase inhibitor, leading to cell cycle arrest and cell apoptosis *in vitro*, and repressing tumor growth and metastasis *in vivo* [27]. Furthermore, Sun et al. showed that in hepatoma cells, Gankyrin downregulation diminished the proportion of cancer stem/progenitor cells and their self-renewal capacity, and this progress occurred partially through regulating hepatocyte nuclear factor 4α (HNF4α), which decided the hepato-

cyte differentiation status and facilitated proteasome-dependent HNF4α degradation [28]. Recently, another study focusing on the effects of Gankyrin on autophagy reported that Gankyrin promoted autophagy in HCC cells in response to starvation or chemotherapy by cytoplasmic interaction with ATG7 and nuclear transactivation of ATG7 expression, demonstrating that Gankyrin might be an attractive target in HCC treatment by inhibiting autophagy and inducing cell sensitivity to chemotherapy [29]. In addition, the oxidative stress status is critical for HCC development and progression, but the mechanisms underlying the manner in which HCC cells respond to excessive oxidative stress remain unclear. In addressing this, Yang et al. found that Gankyrin manipulated HCC cellular redox homeostasis through the feedback regulation of Nrf2 [30]. A recent study reported that Gankyrin expression was markedly reduced by arctigenin (which was an active natural component of Chinese herbal medicine) through inhibiting Gankyrin promoter activity in HepG2 cells, resulting in the repression of HCC cell growth, invasion and metastasis, indicating the importance of Gankyrin as a therapeutic target in HCC [31].

To date, there have been many studies on Gankyrin in HCC, and the overexpression of Gankyrin contributes to HCC carcinogenesis and progression through diverse mechanisms. Thus, it is assumed that targeting Gankyrin may be a rational strategy for HCC treatment, and a thorough understanding of the functions of Gankyrin will be crucial to inform further steps aimed at developing a Gankyrin-based therapy.

Role of Gankyrin in other cancers

Glioma

Gankyrin was overexpressed in glioma compared with para-cancerous tissues and correlated with a high Karnofsky performance score (KPS), advanced WHO grade, and poor prognosis. In addition, U251 cell proliferation was significantly repressed by silencing Gankyrin both *in vitro* and *in vivo*, indicating that Gankyrin might be a potential therapeutic target for glioma treatment [32]. These data are consistent with a role for Gankyrin as an oncogene in glioma; however, unfortunately, specific mechanisms underlying Gankyrin-mediated glioma carcinogenesis and progression have not been investigated, which need for further researches.

Oral cancer

Li et al. examined Gankyrin mRNA and protein expression in human oral epithelial cell lines, at-risk normal oral tissues, premalignant oral lesions, and primary oral squamous cell cancers (OSCCs). Their results demonstrated that both Gankyrin mRNA and protein were overexpressed in 1 premalignant squamous epithelium cell line, 5 malignant OSCCs and the majority of OSCC tissues. *In vitro* study also found that Gankyrin overexpression promoted the proliferation of premalignant and malignant oral cancer cells; by contrast, the downregulation of Gankyrin with siRNA in these cell lines inhibited cell proliferation [33]. Moreover, Gankyrin overexpression may be an early event occurring during oral carcinogenesis, and these findings provide novel insights into the early molecular markers during OSCC development. OSCC is an aggressive disease with a tendency toward relapse and distal metastasis; but whether Gankyrin expression correlates with OSCC metastasis, patient prognosis, radiation therapy and chemotherapy remains unknown.

Lung cancer

Microarray analysis revealed that the levels of Gankyrin expression were much higher in human lung cancer specimens, especially adenocarcinomas, which had frequent *Ras* mutations. In addition, Gankyrin downregulation strongly impaired AKT activation and restrained the carcinogenesis of lung cancer cells that had *Ras* mutations [16]. These results revealed the functional role of Gankyrin and its regulated PI3K/AKT cascade in tumorigenesis induced by oncogenic Ras in lung cancer. Consistent with these findings, Wang et al. found that Gankyrin was overexpressed in non-small lung cancer (NSCLC) compared with the corresponding normal specimens at both the mRNA and protein levels. Gankyrin expression was significantly associated with lymphatic metastasis and TNM stage. Moreover, Kaplan-Meier survival analysis showed a correlation between the overexpression of Gankyrin and poor outcomes for NSCLC patients; presumably, Gankyrin overexpression might serve as an independent prognostic factor in NSCLC [34]. Furthermore, the

authors of that study also demonstrated that Gankyrin could promote epithelial-mesenchymal transition (EMT) and metastasis by regulating IL-6/STAT3 and TGFβ/SMAD3 signaling pathways [35]. Collectively, these data provide novel strategies that can be applied to target Gankyrin and the Gankyrin-related pathways that are important for tumor cell growth, proliferation, and metastasis.

Esophageal cancer

Gankyrin expression was higher in esophageal cancer (ESCC) tissues and 11 ESCC cell lines than in the corresponding normal tissue samples and normal esophageal epithelial cells. In addition, the expression level of Gankyrin was negatively correlated with clinical stage, the poor survival of patients and distant lymph node metastasis. Furthermore, downregulation of Gankyrin with a shRNA vector in KYSE 170 cells repressed cell growth, motility, invasiveness *in vitro* and tumor formation *in vivo* [36]. Moreover, Tang et al. evaluated the role of Gankyrin in ESCC metastasis by measuring Gankyrin mRNA and protein levels in ESCC tissues and cell lines. The results demonstrated that not only Gankyrin mRNA but also Gankyrin protein expression levels were dramatically increased in metastatic ESCC specimens and invasive cell lines compared with normal tissue and normal esophageal epithelial cells. Consistent with the findings of a previous study [36], Gankyrin expression was found to be correlated with T-stage, lymph node metastasis and lymphatic invasion. Functionally, the knockdown of Gankyrin with siRNA in invasive ESCC cells significantly inhibited cell metastasis [37]. These findings raise the possibility that Gankyrin is an important driver for ESCC tumor progression and metastasis, and may be a potentially important therapeutic target in ESCC [36, 37].

Gastric cancer

Liu et al. reported that the regulation of miR-505 by SNP rs111638916 resulted in the elevation of Gankyrin expression levels in gastric cancer patients with GA and AA genotypes, who presented with larger tumors and a higher risk of metastasis [38]. It was speculated that SNP rs111638916 in the Gankyrin gene 3'-UTR was a risk factor for gastric cancer. Another study found that Gankyrin expression was increased in gastric cancer specimens compared with the corresponding normal specimens, and patients with high levels of Gankyrin had a worse overall survival than those with low levels, suggesting that Gankyrin served as a predictive marker for patient prognosis [39]. Furthermore, Gankyrin downregulation strengthened the sensitivity of SGC7901/VCR (vincristine-resistant gastric adenocarcinoma cell line) cells to anticancer drugs and reduced the capacity of cells to efflux adriamycin [40]. Consistent with these findings, Zeng et al. also found that compared to noncancerous tissues, gastric cancer tissues had significantly higher levels of Gankyrin; downregulation of Gankyrin inhibited cancer cell growth and proliferation, while enhanced cell chemosensitivity to 5-fluorouracil and cisplatin was achieved by regulating cell cycle-related proteins, such as cyclin D1, cyclin E and by activating the PI3K/AKT pathway [41]. These studies suggest that Gankyrin plays a significant role in gastric tumorigenesis and may be a predictive marker for prognosis in gastric cancer patients; moreover, its overexpression contributes to the multidrug resistance of gastric cancer cells, and thus, it could be a potentially effective target for chemotherapy.

Colorectal cancer

Tang et al. reported that Gankyrin expression was clearly increased in colorectal cancer (CRC) tissues compared with controls; in addition, clinicopathological analyses showed that Gankyrin overexpression was correlated with TNM stage and metastasis in CRC [42]. Moreover, patients with positive Gankyrin expression had poorer survival than those without Gankyrin expression, indicating that Gankyrin may be valuable in predicting the outcome for CRC patients [43]. When overexpressed in CRC cells (Lovo) with the PhkiNeo-hGankyrin plasmid, Gankyrin promoted cell proliferation and tumorigenesis, whereas RNAi-mediated silencing of Gankyrin expression exerted the opposite effects on CRC SW620 cells [42]. Furthermore, Gankyrin was implicated in cancer metastasis; the expression of Gankyrin was higher in primary CRC with hepatic metastasis than in CRC without metastasis, and an *in vivo* study found the ability of the CRC cells to migrate, invade and metastasize was impaired by silencing Gankyrin [44]. Moreover, IL-8 and cyclin D1 protein levels were positively related to Gankyrin expression levels in CRC tissues. Cyclin D1

deletion was required for Gankyrin-mediated cell migration, and recombinant IL-8 administration rescued the migratory defect in CRC cells. Mechanistically, Gankyrin activated the IL-8 signaling pathway to facilitate the hepatic metastasis of CRC [44]. Recent studies have found that Gankyrin promotes colorectal carcinogenesis and progression by regulating mTORC1 signaling and the PI3K/GSK-3β/β-catenin pathway, indicating that Gankyrin is a potential therapeutic target for the clinical treatment of CRC [45, 46]. Sakurai et al. analyzed the role of Gankyrin in the development of colitis-associated cancer (CAC), results indicated that Gankyrin expression was increased in inflammatory cells and tumor cells in the colonic mucosa of patients with CAC compared with the colonic mucosa of healthy individuals [47]. By binding to SHP-1, Gankyrin activated STAT3 and increased TNF-α and IL-7 levels, leading to the enhancement of inflammation [47]. Collectively, these findings indicate that downregulation of Gankyrin may be a potentially effective strategy for advanced treatment of patients with CAC.

Pancreatic cancer

Meng et al. examined Gankyrin mRNA and protein expression in patients with pancreatic cancer. Both Gankyrin RNA and protein expression levels were significantly higher than those in noncancerous specimens [48]. Furthermore, Gankyrin upregulation could enhance cell proliferation and growth both *in vitro* and *in viv*o, and the downregulation of Gankyrin suppressed cyclin A, cyclin D1, cyclin E, CDK2, CDK4, PCNA and p-Rb but upregulated p27, Rb and p53. These data suggest that Gankyrin contributes to pancreatic cancer cell proliferation through promoting cell cycle progression and p53 degradation [48]. However, the role of Gankyrin in the prediction of pancreatic cancer prognosis is unknown, pending evidence from further researches.

Liposarcoma

Liposarcoma (LPS) is the most common type of soft tissue sarcoma [49]. Gankyrin was demonstrated to be upregulated in both well-differentiated liposarcoma (WDLPS) and dedifferentiated liposarcoma (DDLPS), and the expression level was slightly higher in DDLPS than in WDLPS. High expression level of Gankyrin correlated with tumor recurrence, metastasis and short overall survival. In addition, the high level of Gankyrin expression was negatively correlated with the expression levels of p53 and p21. Through the inhibition of p53 and p21, as well as the AKT/mTOR pathway, DDLPS cell proliferation, colony-formation, migration and tumorigenesis were all inhibited by the downregulation of Gankyrin [50]. Although further studies are needed to clarify the mechanisms by which Gankyrin affects LPC tumorigenesis and promotion, these results suggest that Gankyrin functions as a new potentially predictive and oncogenic factor in WDLPS and DDLPS.

Cervical cancer

Early detection of cervical cancer has recently been a popular topic. Liu et al. reported that Gankyrin expression was elevated in CIN II-III and squamous cancer cell tissues compared with CIN I and benign cervical tissues, suggesting that Gankyrin may predict high-risk disease [51]. Interestingly, nuclei Gankyrin expression was negatively correlated with lymph node metastasis. Moreover, it was reported that Gankyrin mediated the process of EMT in cervical cancer, and the transfection of Gankyrin into cervical SiHa and Hela cancer cells upregulated Vimentin, β-catenin and Twist2 and downregulated E-cadherin, which were important for EMT [51]. In conclusion, although Gankyrin overexpression predicts patients with high risk cervical lesions, the correlation between its expression and cervical cancer patients' survival needs further study.

Ovarian cancer

Ovarian cancer is one of the most lethal gynecological malignancies, and most patients are diagnosed at advanced stages; additionally, the prognosis of these patients remains poor [52]. Therefore, novel biomarkers for the early diagnosis and treatment of ovarian cancer are urgently needed. Gankyrin was significantly overexpressed in ovarian cancer tissues versus benign tumors. Clinicopathological analysis revealed that Gankyrin expression was positively associated with clinical stage and serum CA125 levels, and negatively related to tumor grade; the overall survival curves and disease-free survival curves showed that high levels of Gankyrin expression were correlated with a poor prognosis and early relapse in ovarian cancer patients [53]. Furthermore, the block-

ade of the PI3K/AKT pathway with LY294002 abolished FSH-induced Gankyrin and cyclin D1 expression. Mechanistically, the knockdown of Gankyrin resulted in significant alterations in the PI3K/AKT pathway, whereas Gankyrin overexpression increased the activation of this pathway, which was related to a reduction in the levels of the PTEN protein and an increase in the level of cyclin D1 [53]. Collectively, these findings suggested that Gankyrin facilitates FSH-induced ovarian cancer cell proliferation by regulating the PI3K/AKT signaling pathway and can serve as a potential prognostic biomarker and therapeutic target for ovarian cancer. As ovarian cancer is prone to recurrence and chemotherapy resistance, whether the inhibition of Gankyrin could improve drug sensitivity needs to be evaluated.

Endometrial cancer

Zhang et al. showed that Gankyrin was overexpressed in endometrial cancer tissues compared with non-cancer tissues; moreover, Gankyrin expression was inversely associated with the pattern of PTEN expression [6]. It was demonstrated that Gankyrin played a significant role in estrogen-driven and GPR30-mediated endometrial cell proliferation signaling via the PTEN/PI3K/AKT pathway, and the depletion of Gankyrin in Ishikawa cells inhibited the E2-induced phosphorylation of AKT. Additionally, GPR30, a member of the seven-transmembrane GPCR family, was involved in mediating the estrogen-induced PTEN/PI3K/AKT pathway through the regulation of Gankyrin. Furthermore, GPR30 downregulation reduced the estrogen-induced elevation of Gankyrin and phosphorylated-AKT protein levels and induced PTEN expression, whereas Gankyrin overexpression partially restored the siGPR30-driven PTEN increase and siGPR30-inhibited phosphorylation of AKT [6]. These results highlighted the novel mechanisms of endometrial cellular proliferation via the GPR30-mediated Gankyrin/PTEN/PI3K/AKT signaling cascade.

Breast cancer

Zhen et al. found that Gankyrin was overexpressed and was strongly associated with lymph node metastasis in breast cancer. The depletion of Gankyrin significantly decreased cancer cell migration and invasion. Knockdown of Gankyrin inhibited Rac1 activity and induced large focal adhesions, whereas its overexpression in breast cancer cells led to increased focal adhesion turnover and cell migration [7]. Additionally, the downregulation of Gankyrin in mouse mammary tumor cells significantly inhibited tumor metastasis to the lung in the tumor xenografts [7]. In accordance with that finding, a study by Kim et al. found Gankyrin was overexpressed in invasive breast carcinomas, and its overexpression was correlated with extensive intraductal cancer in invasive ductal cancer; however, there was no association between Gankyrin expression and tumor size, tumor stage, lymphatic or perineural invasion, or lymph node metastasis. In addition, Gankyrin downregulation reduced cell proliferation and tumorigenesis in MCF7 breast cancer cells [54]. Hypoxia is critical for tumor growth and metastasis. Gao et al. reported that Gankyrin deletion impaired the increased migration and invasion of BT474 breast cancer cells under hypoxic settings partly through regulating E-cadherin, highlighting that Gankyrin might promote tumor metastasis through the regulation of Rac1 and E-cadherin activity [55]. Collectively, these data demonstrate that Gankyrin plays essential roles in breast cancer, and a comprehensive understanding of the molecular aberrations of Gankyrin may help in the development of new targeted agents.

Prognostic value of Gankyrin in human cancer

Gankyrin levels are progressively elevated as normal tissues become tumors, making the level of Gankyrin a promising marker for cancer diagnosis, but the prognostic value of Gankyrin in cancer remains unknown. As indicated above, although most studies found that high levels of expression of Gankyrin were significantly associated with poor prognoses in a variety of human malignancies, unfortunately, whether Gankyrin overexpression could predict poor outcomes in HCC patients was controversial and needed for further study [4, 5, 25]. Recently, a meta-analysis including 1,326 cancer patients was undertaken to investigate whether Gankyrin overexpression predicted a worse prognosis. Gankyrin overexpression was associated with poorer overall survival with a hazard ratio (HR) of 1.73 and a 95% confidence interval (CI) of 1.29-2.31. Moreover, in Chinese patients, particularly in patients with digestive system cancers, the overexpression of Gankyrin was significantly related to advanced TNM sta-

ge (RR=0.72, 95% CI: 0.60-0.86), positive lymph node metastasis (RR=0.72, 95% CI: 0.60-0.86) and distant metastasis (RR=1.43, 95% CI: 1.20-1.70), suggesting that Gankyrin may be a promising biomarker for the prediction of clinical outcomes for such patients [56]. However, the relationship between Gankyrin level and tumor prognosis should be validated by multiple researches with large sample sizes.

Conclusions and perspectives

Overall, Gankyrin plays an important role in a variety of human malignant tumors, in addition to its roles in tumors described above, Gankyrin is also upregulated in some other human cancers, such as cholangiocarcinoma [17, 21], testicular germ cell tumor [57], and prostate cancer [58], functioning as an oncogene and influencing tumor cell growth. Tumor heterogeneity remains a major hurdle for cancer treatment; the evaluation of the molecular events underlying tumorigenesis would be helpful for the development of precisely diagnostic and therapeutic modalities. In the era of personalized cancer medicine, the identification of molecular genetic biomarkers has considerably improved our current knowledge of cancer diagnosis, prognosis, and therapeutic strategies. Emerging evidence has demonstrated that aberrant genetic changes play important roles in the multiple-step processes of tumorigenesis and tumor progression. A large number of studies have demonstrated Gankyrin may be a promising target for future therapies in a broad range of tumor types. Firstly, Gankyrin can act as a molecular biomarker for cancer diagnosis, and the overexpression of Gankyrin predicts tumor progression and promotion in various human malignancies. Secondly, downregulation of the Gankyrin gene can inhibit tumor growth and metastasis, so it may be an attractive option for cancer management. Finally, Gankyrin may help to identify responders who are likely to benefit from chemotherapy.

While most studies point to a direct relationship between Gankyrin overexpression and tumor progression, studies about Gankyrin in tumor biology have been far from complete. Therefore, more efforts are still needed to characterize the comprehensive molecular traits of Gankyrin before transforming the experimental studies of Gankyrin into clinical applications.

Acknowledgements

This work was supported by Beijing Natural Science Foundation (No. 7184206).

Disclosure of conflict of interest

None.

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