Cancer Letters 451 (2019) 150–155

Contents lists available at ScienceDirect

Cancer Letters

journal homepage: www.elsevier.com/locate/canlet

Mini-review

# Card9 as a critical regulator of tumor development

Xiaoming Zhong[b,1], Bin Chen[c,1], Liang Yang[d], Zhiwen Yang[a,*]

[a] Department of Pharmacy, Songjiang Hospital Affiliated Shanghai First People's Hospital, Shanghai Jiao Tong University, Shanghai, China
[b] Jiangxi Province Tumor Hospital, Nanchang, China
[c] Surgery Department, First Affiliated Hospital of Gannan Medical University, Gannan Medical University, Ganzhou, China
[d] Nanjing Medical University, The Affiliated Changzhou No.2 People's Hospital, Nanjing Medical University, Nanjing, China

ARTICLE INFO

*Keywords:*
Card9
Macrophages
Tumor growth
Target therapy

ABSTRACT

Caspase recruitment domain-containing protein 9 (Card9) is a myeloid cell-specific signaling protein that plays a critical role in NF-κB and MAPK activation. This leads to initiation of the inflammatory cytokine cascade, and elicits the host immune response against microbial invasion, especially in fungal infection. Current research indicates that Card9 plays an important role in tumor progression. Here, we review the data from preclinical and clinical studies of Card9 and suggest the potential for Card9-targeted interventions in the prevention or treatment of certain tumors.

## 1. Introduction

Caspase recruitment domain-containing protein 9 (Card9) is a central integrator of innate and adaptive immunity that is mainly expressed in myeloid cells, especially in macrophages and dendritic cells. Card9 has been identified as the downstream effector molecule of the pattern recognition receptors in myeloid cells. Following receptor engagement, Card9, as an intracellular adaptor molecule, can activate the NF-κB and/or MAPK signaling pathways, leading to an inflammatory cascade against invasive bacteria, fungi and viruses [1–6]. Thereby, *Card9*, an inflammation-related gene, has been extensively studied in terms of its potential involvement in infectious inflammatory diseases.

Understanding the factors that contribute to the initiation of cancer development will be crucial in fighting cancer [7,8]. Recently, increasing evidence has emerged that Card9 may play an important role in multiple aspects of cancer biology [9,10]. In this review, we summarize the clinical data concerning Card9 expression and outcomes in tumor patients, discuss the profound influence of Card9 signaling throughout each step of carcinogenesis, and highlight the potential utility in manipulating Card9 signaling for tumor suppression.

## 2. Clinical significance of Card9 in tumor patients

### 2.1. Hepatocellular carcinoma

Hepatocellular carcinoma (HCC) is one of the most lethal cancers in humans. Among the various etiological factors, hepatitis C virus (HCV) infection is a major cause of HCC [11]. It is of great value for understanding the HCC progression in HCV-infected patients.

Zekri and colleagues studied 130 patients with HCV-associated liver disease, 40 of which were diagnosed with HCV-associated HCC [12]. To address the role of Card9 in HCC patients, Card9 mRNA in the liver was quantitatively estimated by RT-PCR. The expression levels of Card9 mRNA in HCV-related HCC were significantly higher than those in the other three groups of HCC, i.e., caused by chronic HCV, chronic active hepatitis, and liver cirrhosis. The best cutoff for Card9, a statistical indicator that can successfully differentiate between HCC and non-HCC, was 47 with 80.0% sensitivity, 80.0% specificity, a 85.7% negative predictive value and a 61% positive predictive value. Therefore, Card9 mRNA could serve as a useful clinical marker for prediction of HCC in HCV-infected patients.

### 2.2. Intestinal carcinoma

Intestinal carcinoma, one of the most prevalent cancers, is the fourth leading cause of cancer death worldwide [13]. To date, the molecular mechanisms underlying cancer growth, invasion and metastasis for intestinal carcinoma remain unknown.

A recent study conducted by Yang et al. indicated a strong clinical correlation between Card9 expression and human colon carcinoma [9]. First, the expression levels of Card9 mRNA and protein were assessed in colon carcinoma patients, showing high expression in tumor tissues

* Corresponding author. Songjiang Hospital Affiliated the First People's Hospital, Shanghai Jiao Tong University, Shanghai, 201600, China.
*E-mail address:* yangzhiwen2009@sina.com (Z. Yang).
[1] These authors contributed equally to this work.

https://doi.org/10.1016/j.canlet.2019.03.001
Received 31 August 2018; Received in revised form 18 February 2019; Accepted 1 March 2019

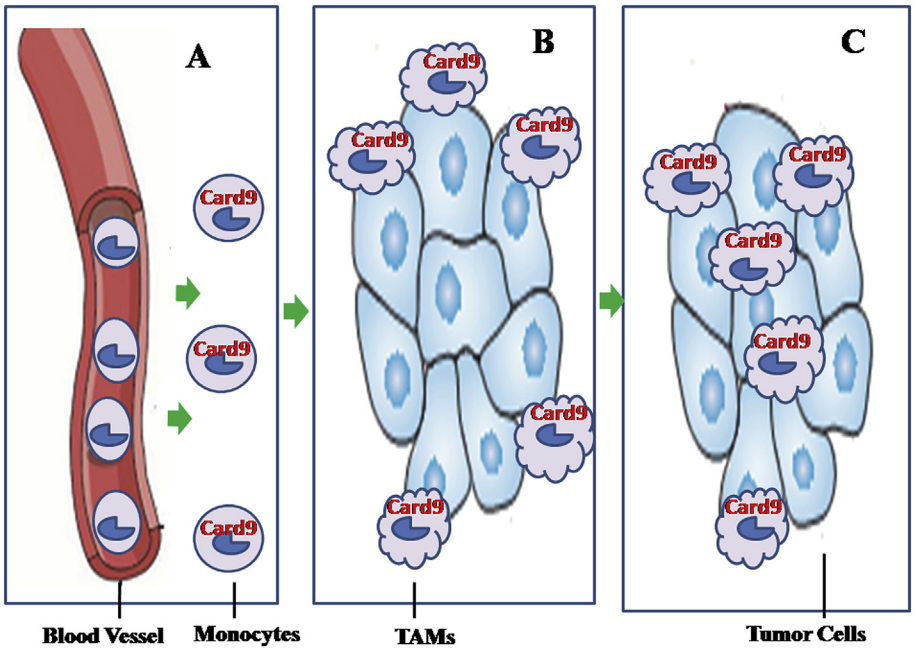


**Fig. 1.** Card9 cellular distribution in intestinal carcinoma. Card9 expression in carcinoma tissues was primarily located in tumor-infiltrating macrophages, not in cancer cells. A: Monocytes from blood vessels were released into tissues; B: Low infiltration of TAMs expressing Card9 into tumor tissues; C: High infiltration of TAMs expressing Card9 into tumor tissues.

compared with normal tissues. Second, the clinicopathologic analysis of 48 cases indicated that Card9 expression was negatively correlated with tumor differentiation and positively correlated with tumor invasion depth and metastasis, further supporting its potential to foster tumor metastasis. Third, immunohistochemical staining demonstrated that increased Card9 expression in colon carcinoma tissues was primarily located in tumor-infiltrating macrophages and not in cancer cells (Fig. 1). The same distribution of Card9 was observed in metastatic lymph nodes from patients. These findings suggested that progression and metastasis of human colon carcinoma may be attributed to aberrant Card9 expression in macrophages.

Enteropathy-associated T-cell lymphoma (EATL) is a rare primary T-cell lymphoma in the human small intestines. In this study, *Card9* was identified as a potential candidate gene for EATL using a high-resolution oligonucleotide microarray, and was subsequently validated in 20 patients by fluorescence *in situ* hybridization. The genomic profile showed that recurrent copy number gains were found at 9q34.13 (Card9). Of note, compared with the other genetic abnormalities in EATL patients, the gain at locus 9q34 (Card9) was the most frequent copy number change (15/20 patients, 75% frequency) [14]. Finally, this study reported a similar frequency of copy number gain at locus 9q34 between Japanese and European EATL cases, irrespective of East and West ethnic differences [15].

### 2.3. Gastric carcinoma

Primary gastric lymphoma is an unusually encountered cancer that originates within the stomach. Approximately 90% of patients with primary gastric lymphoma possess mucosa-associated lymphoid tissue (MALT) gastric lymphoma and diffuse large B-cell lymphoma (DLBCL). MALT gastric lymphoma is often linked to infection with the *Helicobacter pylori* bacterium in the gastric mucosa [16].

Two studies were reported that investigated the correlations between Card9 mRNA expression and clinical characteristics in patients with gastric carcinoma [17,18]. In the first study, 65 patients were diagnosed with primary gastric lymphoma, of which 43 patients had low-grade MALT lymphoma, 16 patients had DLBCL plus MALT lymphoma, and 6 patients had DLBCL without MALT lymphoma. The results showed a positive detection rate for Card9 mRNA of up to 48% in tissue specimens from the 65 patients. However, there was no statistically significant difference in Card9 mRNA between MALT lymphomas and DLBCL (positive rates 46% vs. 67%, $P > 0.05$). However, a higher positive frequency of Card9 mRNA was detected in *H. pylori*-negative patients than in *H. pylori*-positive patients. These findings suggested that Card9 was associated with primary gastric lymphoma, especially in *H. pylori*-negative patients [17]. In the second study, a total of 26 patients who suffered from gastric MALT lymphoma with or without t(11; 18) (q21; q21) were included. Discrete recurrent chromosomal gains at locus 9q34 (Card9) were found to be a common feature in t(11; 18)-negative gastric MALT lymphoma, but rarely observed in those positive for this chromosomal translocation. It was therefore concluded that Card9 contributes to the pathogenesis of t(11; 18)-negative MALT lymphoma [18].

### 2.4. Kidney carcinoma

Clear cell renal cell carcinoma (ccRCC) is the most common type of kidney cancer, responsible for approximately 80% of all renal neoplasms [19]. Recently, Tan and co-workers identified Card9 as a potential biomarker for ccRCC risk and clinical outcomes. Card9 expression variation between tumor and adjacent normal tissues was detected in the two populations with ccRCC, the 93 patients in the screened population and the 258 patients in the validated population. After careful analysis of the clinical outcomes in the 258 patients in the validation population, Card9 overexpression was found to be detrimental to ccRCC patients, as indicated by a 2.11-fold increase in the risk of death. This was the first report of a correlation between Card9 expression and clinical outcomes in tumor patients [20].

### 2.5. Malignant pleural effusion

Malignant pleural mesothelioma is defined as an aggressive tumor with a poor prognosis. In current clinical practice, there is no highly sensitive and specific tumor biomarker for malignant pleural effusion [21]. In this study, a total of 143 patient samples, including 83 cases of malignant pleural effusion and 60 cases of benign pleural effusion, were collected to establish the protein profiles using matrix-assisted laser desorption/ionization time-of-flight mass spectrometry. Compared with benign pleural effusion, Card9 (3930.9 $m/z$) was obviously decreased in malignant pleural effusion. A possible reason for this was related to the selected control group. If any of the healthy volunteers in the control group had pleural effusion, relative Card9 expression in the tumor

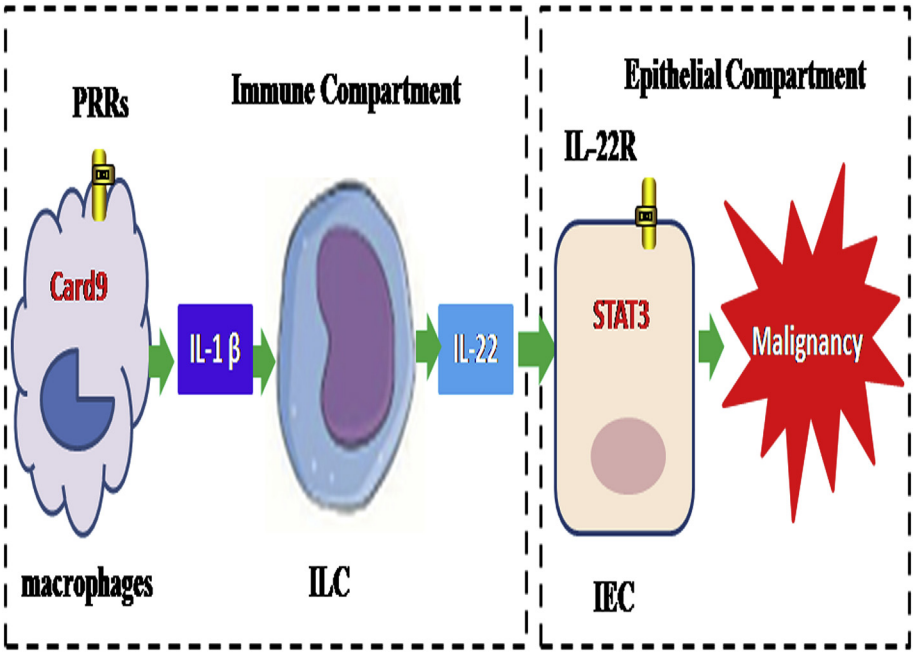


**Fig. 2.** Card9 function promoted tumor growth indirectly through the regulation of immune cells. Card9 was not expressed within intestinal epithelial cells (IEC) but specifically in infiltrated macrophages. Card9 specifically regulated IL-22 production in an IL-1β-dependent manner in type 3 innate lymphoid cells (ILC), leading to intrinsic STAT3 activation in IEC, and eventually inducing malignancy and colitis-associated cancer.

patients would be increased. Interestingly, Card9 could serve as a useful peptide biomarker for the diagnosis of malignant pleural effusion, with 89.6% sensitivity, 88.2% specificity, a 92.8% positive predictive value, a 83.3% negative predictive value and 89.1% accuracy [22].

As is the case with intestinal carcinoma, for most tumor patients with malignant pleural effusion it was not determined whether Card9 protein originated from tumor-infiltrating macrophages or cancer cells. As Card9 is a myeloid cell-specific signaling protein, it would be expected to originate from macrophages. However, in-depth analyses indicated that Card9 is not only expressed in macrophages but also in tumor cells. Thereby, it could not be completely excluded from tumor cells.

## 3. Preclinical studies of Card9 in tumors

### 3.1. Tumor growth

In the mouse model of colitis-associated cancer, the innate immune adapter Card9 triggered a vigorous host immune response against tumor growth (Fig. 2) [23]. The deletion of Card9 in mice could influence the regeneration of intestinal epithelial cells, induce cellular apoptosis, develop a lower degree of dysplasia, inhibit the proliferation of tumor cells, and reduce polyp size, but did not alter the total number of neoplasms per mouse. This evidence suggested an obvious inhibition of colitis-associated tumor growth, instead of tumor initiation, in Card9-deficient mice. Importantly, Card9 was not expressed within intestinal epithelial cells but specifically in infiltrated macrophages, revealing Card9 as a signal amplifier for macrophage activation in intestinal tumors. Intriguingly, complete inhibition of IL-1β and IL-17A expression, and a significant reduction in IL-22 expression, was observed within colonic lamina propria macrophages from Card9-deficient mice. IL-1β, which is a key growth factor for intestinal epithelial cells, not only regulates Th17 cell responses, but also maintains and enhances the production of IL-22 by innate lymphoid cells [24–26]. What is more, Card9 specifically regulates IL-22 production in an IL-1β-dependent manner in type 3 innate lymphoid cells, leading to intrinsic STAT3 activation. IL-22 acted effectively on epithelial cells in the promotion of dysplasia and tumor growth via STAT3 activation [27]. Thus, Card9 signaling predisposed to tumor growth in colitis-associated cancer.

### 3.2. Tumor promotion

In the $APC^{min}$ mouse model, Card9 promoted tumorigenesis in sex-biased colon tumors, specifically in male mice [28]. As is well-known, the $APC^{min}$ mouse model mimics the genetic lesions associated with human familial adenomatous polyposis. Tumor multiplicity was obviously decreased in Card9-deficient male mice, while average tumor size remained unchanged, confirming significant promotion of tumorigenesis but not of tumor growth. Subsequently, enhanced viability was found in male Card9-deficient mice. Sex-biased colon tumors may therefore be attributed to decreased plasma cytokines (IL6, G-CSF and RANTES), and less T-cell and macrophage infiltration into colonic tumor tissues. A previous report showed that factors upstream of plasma IL6 enhanced cachexia and reduced viability in male $APC^{min}$ mice but not female $APC^{min}$ mice [29]. Further studies will be required to test the sex-biased role of Card9-induced IL6 and Th17-cell in colon tumorigenesis.

### 3.3. Tumor metastasis

In colon carcinoma-bearing mice, Card9 in macrophages plays a critical role in facilitating liver metastasis of colon carcinoma cells (Fig. 3) [9]. In this study, higher levels of antitumor cytokine IL-12 and lower levels of tumor-promoting cytokine IL-10, IL-1α, transforming growth factor-β, and vascular endothelial growth factor receptor 1, were found in $Card9^{-/-}$ mice. Consistent with the cellular distribution reported previously, Card9 was abundantly expressed in tumor-infiltrating macrophages rather than tumor cells. Due to the lack of Card9 expression in tumor-infiltrating macrophages, tumor metastasis in $Card9^{-/-}_{BM}$→WT mice was obviously inhibited compared with $WT_{BM}$→WT mice. Importantly, the proportion of CD206-positive macrophages was predominant in the hepatic metastatic tumors of WT mice, while the population of M2 macrophages (CD206 + cells) was rarely detected in the metastatic foci of Card9-deficient mice. CD206 is a specific marker of M2 macrophages [30]. These results suggested that tumor cell-activated Card9 signaling contributed to macrophage polarization toward the metastasis-promoting phenotype responsible for tumor metastasis. To further determine the possible mechanisms by which tumor cells modulate macrophage polarization, tumor cell-secreted vascular endothelial growth factor (VEGF) was identified. It is well

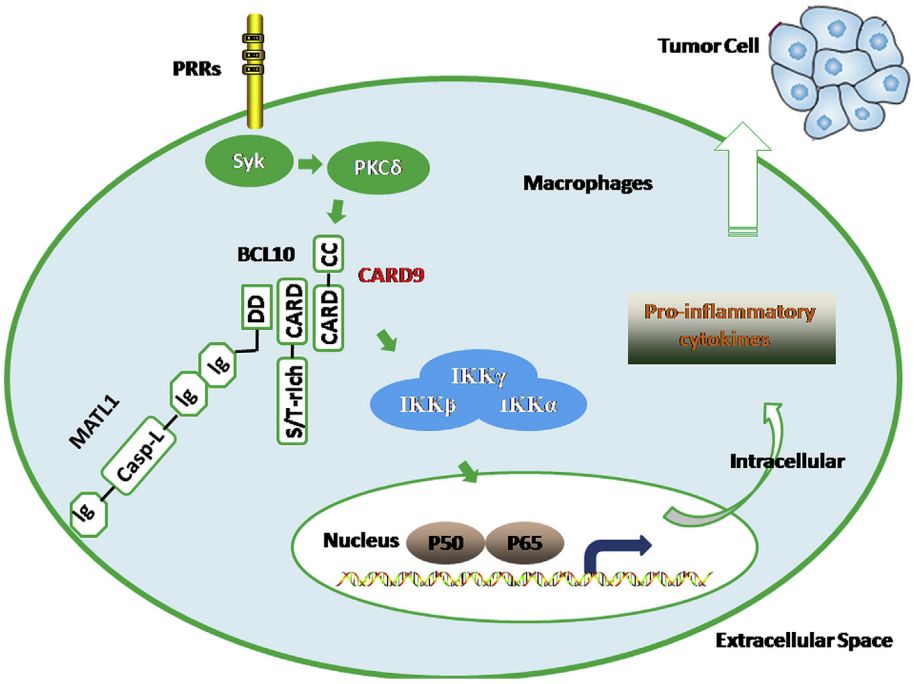


**Fig. 3.** Card9 function supported tumor growth directly by the secretion of cytokines and growth factors. Card9 was expressed in infiltrating macrophages. Ligand recognition by pattern recognition receptors (PRRs) leads to the recruitment of spleen tyrosine kinase (Syk), the activation of protein kinase C (PKC) δ and the formation of the CARD9/BCL10/MATL1(CBM) complex. The CBM complex regulated NF-κB signaling, which led to the secretion of pro-inflammatory factors, and facilitated liver metastasis of colon carcinoma cells.

known that VEGF protein secreted by tumor cells under hypoxic conditions can promote tumor cell invasion and metastasis [31]. Of note, this study found that VEGF in the tumor microenvironment could induce macrophage Syk activity in a paracrine manner. Subsequently, Syk phosphorylation in macrophages stimulated the intracellular assembly of the CARD9–BCL10–MALT1 complex, leading to the activation of NF-κB signaling and eventually driving macrophage polarization toward the metastasis-promoting phenotype.

### 3.4. Tumor immunity

Cross-priming refers to a process by which antigen-presenting dendritic cells present exogenous antigens in the context of MHC class I (MHC-I) for the activation of $CD8^+$ cytotoxic T lymphocytes (CTLs) [32]. The cross-priming process plays a critical role in antiviral and antitumor immune responses.

It was recently reported that Card9 activation in dendritic cells could potently cross-prime antigen-specific $CD8^+$ CTLs with long-lasting antitumor responses [33]. As described previously, the Dectin-1 agonist Curdlan serves as an exogenous antigen to activated dendritic cells in the Card9-dependent pathway [1], and OVA is an octameric peptide from ovalbumin presented by MHC-I [34]. As inducers of all arms of adaptive immunity, Dectin-1 ligands are interesting candidates to enhance vaccine strategies, and have been used successfully to trigger antigen-specific T-cell responses [35]. After treatment with Curdlan and OVA, one study confirmed that Card9, as a central adapter molecule, enhanced and improved Dectin-1-mediated cross-priming of $CD8^+$ CTLs. What is more, the cross-priming function was largely abrogated in Card9-deficient dendritic cells and rat models. As reported previously, Dectin-1 signaling in dendritic cells could induce NK cell-mediated immune responses to tumor cell killing [36]. This raised the question as to whether NK cells may play an active role in the antitumor response. Fortunately, Card9 targeted depletion demonstrated that the Dectin-1-induced antitumor immune response against melanoma was independent of NK cell function, which exclusively relied on $CD8^+$ CTLs in this study. As a result, Card9-deficient mice failed to control melanoma growth or prolong survival following treatment with OVA and Curdlan [33].

### 3.5. Tumor cell apoptosis

*Card9* is also an important apoptosis-inducing gene in leucocythemia [37,38]. In a previous study, the human acute promyelocytic leukemic cell line NB4 was used to explore the potential role of Card9. The *SALL4* gene is known to function in the control of cell proliferation and apoptosis. After *SALL4* knockdown, the aberrant expression of apoptosis genes (encoding caspase-3, annexin V, and DNA fragmentation proteins) was found in NB4 cells, leading to the promotion of cell apoptosis and decreased tumorigenicity. Most striking, quantitative real-time PCR established that Card9 was significantly upregulated along with a reduction in SALL4 expression. Chromatin immunoprecipitation validated that SALL4 could bind to the promoter of the *Card9* gene, indicating that *Card9* is a downstream target of SALL4. Thereby, the *Card9* gene was responsible for SALL4-mediated leukemogenesis through the induction of cell apoptosis [37]. To further analyze the role of Card9 as a key regulator in leukemic cell apoptosis, the human promyelocytic cell line HL60 was used as another cellular model. In this study, HL60 leukemic cells were incubated with arsenic trioxide alone and in combination with bortezomib. After drug treatment, the upregulation of some proapoptotic genes (*Card9*) and the downregulation of some antiapoptotic genes were observed with bortezomib, but not with arsenic trioxide alone. However, the molecular and cellular functions of Card9 were not reported in this study [38]. Previous research exhibited that arsenic trioxide and bortezomib with strong antileukemic activity inhibit cell proliferation and induce apoptosis through the NF-κB signaling pathway [39,40]. Increasing evidence has shown that NF-κB transcriptional activity is dependent on Card9 [1]. These findings suggested that Card9 may be involved in HL60 cell apoptosis.

### 3.6. Card9-dependent signaling pathway in tumorigenesis

The Card9-dependent NF-κB and JNK molecular signaling pathways are reported to drive tumor growth of VHL tumor suppressor protein (pVHL)-deficient ccRCC [10,41]. The pro-tumor mechanisms of Card9 in kidney tumor cells can be attributed to cytokines, chemokine release, and epithelial–mesenchymal transition.

It was shown that pVHL, which bound to casein kinase 2, promoted

**Table 1**
Preclinical studies of Card9 in tumors.

| Card9 Function | Tumor | Mechanisms | References |
|---|---|---|---|
| Promotion | Intestine | IL-1β, IL-17A, IL-22, STAT3 | [19] |
| Promotion | Intestine | IL6,G-CSF,RANTES,T-cell, macrophage | [24] |
| Promotion | Intestine | IL-12,IL-10,IL-1α,TGF-β,NF-κB, M2 macrophage | [5] |
| Promotion | Kidney | Card9 linked pVHL to NF-κB biology | [37] |
| Promotion | Kidney | Card9-induced JNK hyper-activation | [6] |
| Inhibition | melanoma | Dectin-1-mediated cross-priming of $CD8^+$ CTLs | [29] |
| Inhibition | leucocythemia | leukemic cell apoptosis | [33,34] |

Transforming growth factor-β (TGF-β), Regulated upon activation normal T cell expressed and secreted factor (RANTES), VHL tumor suppressor protein (pVHL), Granulocyte colony-stimulating factor (G-CSF), c-Jun N-terminal kinase (JNK), Cytotoxic T lymphocytes (CTLs).

the phosphorylation of Card9 at the C terminus, impairing its ability to activate NF-κB. In the context of VHL loss, there was constitutive activation of Card9, an upstream molecule of NF-κB signaling, and elimination of Card9 normalized NF-κB activity. Consequently, down-regulation of Card9 in $pVHL^{-/-}$ cancer cells restored their sensitivity to the cytokine TNF-α and retarded tumor growth. Therefore, Card9 is the link between pVHL and NF-κB signaling in the control of ccRCC [41].

Card9-induced JNK hyper-activation also played a critical role in pVHL-deficient ccRCC [10]. In the context of pVHL inactivation, the inhibition of Card9 phosphorylation by CK2 allowed for the formation of a protein complex involving CARD9, BCL10, and TRAF6. Then, TRAF6 ubiquitination through K63 linkages upstream triggered the sequential functions of TAK1 and JNK. Conversely, Card9 RNA interference led to a sharp reduction in TRAF6 K63 polyubiquitination, inhibiting the formation of a CARD9–TRAF6 complex, and eventually impairing the activation of the JNK signaling axis. Of note, pVHL-positive cells constitutively expressed higher levels of Card9 compared with pVHL-negative cells, but CARD9–TRAF6 interactions were formed more robustly in pVHL-deficient cells. To our knowledge, the formation of a stable complex between Card9 and other proteins, triggered its biological functions [1]. As a result, the CARD9/TRAF6 complex mediated the association between pVHL and JNK in ccRCC.

## 4. Card9 as a target for tumor therapy

It has been extensively documented that as a natural product β-glucans contribute to immune cell responses by stimulating their anti-tumor activity [42]. In this study, the possible mechanism by which β-glucan treatment regulated these processes was investigated. β-glucan treatment converted tumor-associated macrophages (TAMs) from an M2-like pro-tumor phenotype into an M1-like anti-tumor phenotype, and enhanced the activation of TAM-induced CD4 and CD8 T cells, leading to reduced tumor progression. In addition, β-glucan selectively binds to cell-surface Dectin-1 receptors, triggering a cascade of Syk kinase, following which Syk is recruited and activates Card9 recruitment to form a Card9/Bcl10 complex that induces Erk phosphorylation. Strikingly, β-glucan-induced Erk phosphorylation was completely abrogated in Card9 knockout mice, indicating that Erk activation was dependent on Card9. Taken together, these data suggested that β-glucan was capable of converting TAMs into an M1-like phenotype through the dectin-1-dependent canonical Syk–Card9–Erk pathway, providing a potential target for antitumor therapy with β-glucan [43].

## 5. Conclusions

Card9 is a central integrator in innate immune cell activation, which triggers the inflammatory signaling pathway in response to microbial infection. Along with an in–depth understanding of pathological Card9 signaling, Card9 was validated to play an essential role in the pathogenesis of tumor growth. In preclinical experimental studies, Card9 signaling was found to promote tumor growth in intestinal and kidney cancer, and suppress tumor growth in melanoma and leucocythemia (Table 1). It generally exhibited pro-tumor functions by activating the STAT3 and NF-κB signaling pathway, and exerted anti-tumor activity by inducing cancer cell apoptosis and generating tumor antigen-specific $CD8^+$ CTL cross-presentation. Future studies will be needed to evaluate its different roles in various tumor models. This discrepancy may be accounted for by its different cellular distribution. The function of Card9 in macrophages and kidney cancer cells plays a pivotal role in facilitating tumor growth, but not in dendritic and leucocythemia cells. It remains to be determined whether Card9 is involved in positive and negative regulation of tumorigenesis in the complicated tumor microenvironment. Clinical studies suggested that the abnormal expression of Card9 was a risk factor for cancer patients, and was associated with tumor progression and poor survival rates. To date, it remains uncertain whether Card9 expression is beneficial to tumor patients. This requires further confirmation using a large cohort of patients in the future.

Despite many uncertainties in this emerging field, recent findings support the fundamental role of macrophages in malignant disease processes. Further studies including preclinical and clinical trials are now required to understand how macrophages interact with tumor cells through Card9 signaling. In the future, Card9-targeted interventions with small molecular inhibitors may be effective in the prevention or treatment of certain tumors.

## Conflicts of interest

The authors report no conflicts of interest in this work.

## Acknowledgement

This study was supported by the Science and Technology Commission of Shanghai Municipality (201840035).

## References

[1] X. Zhong, B. Chen, L. Yang, Z. Yang, Molecular and physiological roles of the adaptor protein CARD9 in immunity, Cell Death Dis. 9 (2018) 52.
[2] X.M. Jia, B. Tang, L.L. Zhu, Y.H. Liu, X.Q. Zhao, S. Gorjestani, et al., CARD9 mediates Dectin-1-induced ERK activation by linking Ras-GRF1 to H-Ras for antifungal immunity, J. Exp. Med. 211 (2014) 2307–2321.
[3] O. Gross, A. Gewies, K. Finger, M. Schafer, T. Sparwasser, C. Peschel, et al., Card9 controls a non-TLR signalling pathway for innate anti-fungal immunity, Nature 442 (2006) 651–656.
[4] S. Roth, A. Rottach, A.S. Lotz-Havla, V. Laux, A. Muschaweckh, S.W. Gersting, et al., Rad50-CARD9 interactions link cytosolic DNA sensing to IL-1beta production, Nat. Immunol. 15 (2014) 538–545.
[5] D. Strasser, K. Neumann, H. Bergmann, M.J. Marakalala, R. Guler, et al., Syk kinase-coupled C-type lectin receptors engage protein kinase C-sigma to elicit Card9 adaptor-mediated innate immunity, Immunity 36 (2012) 32–42.
[6] Z. Cao, K.L. Conway, R.J. Heath, J.S. Rush, E.S. Leshchiner, Z.G. Ramirez-Ortiz, et al., Ubiquitin ligase TRIM62 regulates CARD9-mediated anti-fungal immunity and intestinal inflammation, Immunity 43 (2015) 715–726.
[7] T. Palaga, W. Wongchana, P. Kueanjinda, Notch signaling in macrophages in the context of cancer immunity, Front. Immunol. 9 (2018) 652.
[8] A. Mantovani, F. Marchesi, A. Malesci, L. Laghi, P. Allavena, Tumour-associated macrophages as treatment targets in oncology, Nat. Rev. Clin. Oncol. 14 (2017) 399–416.
[9] M. Yang, J.H. Shao, Y.J. Miao, W. Cui, Y.F. Qi, J.H. Han, et al., Tumor cell-activated CARD9 signaling contributes to metastasis-associated macrophage polarization,

Cell Death Differ. 21 (2014) 1290–1302.
[10] J. An, H. Liu, C.E. Magyar, Y. Guo, M.S. Veena, E.S. Srivatsan, et al., Hyperactivated JNK is a therapeutic target in pVHL-deficient renal cell carcinoma, Cancer Res. 73 (2013) 1374–1385.
[11] S.B. Prenner, L.B. VanWagner, S.L. Flamm, R. Salem, R.J. Lewandowski, L. Kulik, Hepatocellular carcinoma decreases the chance of successful hepatitis C virus therapy with direct-acting antivirals, J. Hepatol. 66 (2017) 1173–1181.
[12] A.R. Zekri, M. El-Kassas, Y. Saad, A. Bahnassy, H.K. El-Din, S.K. Darweesh, et al., Caspase recruitment domains. New potential markers for diagnosis of hepatocellular carcinoma associated with HCV in Egyptian patients, Ann. Hepatol. 12 (2013) 774–781.
[13] R. Lozano, M. Naghavi, K. Foreman, S. Lim, K. Shibuya, V. Aboyans, et al., Global and regional mortality from 235 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the Global Burden of Disease Study 2010, Lancet 380 (2012) 2095–2128.
[14] S. Tomita, Y.Y. Kikuti, J. Carreras, M. Kojima, K. Ando, H. Takasaki, et al., Genomic and immunohistochemical profiles of enteropathy-associated T-cell lymphoma in Japan, Mod. Pathol. 28 (2015) 1286–1296.
[15] R.J. Deleeuw, A. Zettl, E. Klinker, E. Haralambieva, M. Trottier, R. Chari, et al., Whole-genome analysis and HLA genotyping of enteropathy-type T-cell lymphoma reveals 2 distinct lymphoma subtypes, Gastroenterology 132 (2007) 1902–1911.
[16] H. Boot, D. de Jong, Diagnosis, treatment decisions, and follow up in primary gastric lymphoma, Gut 51 (2002) 621–622.
[17] S. Nakamura, S. Nakamura, T. Matsumoto, S. Yada, M. Hirahashi, H. Suekane, et al., Overexpression of caspase recruitment domain (CARD) membrane-associated guanylate kinase 1 (CARMA1) and CARD9 in primary gastric B-cell lymphoma, Cancer 104 (2005) 1885–1893.
[18] Y. Zhou, H. Ye, J.I. Martin-Subero, R. Hamoudi, Y.J. Lu, R. Wang, et al., Distinct comparative genomic hybridisation profiles in gastric mucosa-associated lymphoid tissue lymphomas with and without t(11;18)(q21;q21), Br. J. Haematol. 133 (2006) 35–42.
[19] S. Chevrier, J.H. Levine, V.R.T. Zanotelli, K. Silina, D. Schulz, M. Bacac, et al., An immune atlas of clear cell renal cell carcinoma, Cell 169 (2017) 736–749.
[20] W. Tan, M.A. Hildebrandt, X. Pu, M. Huang, J. Lin, S.F. Matin, et al., Role of inflammatory related gene expression in clear cell renal cell carcinoma development and clinical outcomes, J. Urol. 186 (2011) 2071–2077.
[21] A. Scherpereel, F. Wallyn, S.M. Albelda, C. Munck, Novel therapies for malignant pleural mesothelioma, Lancet Oncol. 19 (2018) 161–172.
[22] H. Li, Z. Tang, H. Zhu, H. Ge, S. Cui, W. Jiang, Proteomic study of benign and malignant pleural effusion, J. Cancer Res. Clin. Oncol. 142 (2016) 1191–1200.
[23] H. Bergmann, S. Roth, K. Pechloff, E.A. Kiss, S. Kuhn, M. Heikenwalder, et al., Card9-dependent IL-1beta regulates IL-22 production from group 3 innate lymphoid cells and promotes colitis-associated cancer, Eur. J. Immunol. 47 (2017) 1342–1353.
[24] K. Sugimoto, A. Ogawa, E. Mizoguchi, Y. Shimomura, A. Andoh, A.K. Bhan, et al., IL-22 ameliorates intestinal inflammation in a mouse model of ulcerative colitis, J. Clin. Investig. 118 (2008) 534–544.
[25] T. Hughes, B. Becknell, A.G. Freud, S. McClory, E. Briercheck, J. Yu, et al., Interleukin-1beta selectively expands and sustains interleukin-22+ immature human natural killer cells in secondary lymphoid tissue, Immunity 32 (2010) 803–814.
[26] G.F. Sonnenberg, L.A. Fouser, D. Artis, Border patrol: regulation of immunity, inflammation and tissue homeostasis at barrier surfaces by IL-22, Nat. Immunol. 12 (2011) 383–390.
[27] S. Kirchberger, D.J. Royston, O. Boulard, E. Thornton, F. Franchini, R.L. Szabady, et al., Innate lymphoid cells sustain colon cancer through production of interleukin-22 in a mouse model, J. Exp. Med. 210 (2013) 917–931.
[28] V.I. Leo, S.H. Tan, H. Bergmann, P.Y. Cheah, M.H. Chew, K.H. Lim, et al., CARD9 promotes sex-biased colon tumors in the APCmin mouse model, Cancer Immunol. Res. 3 (2015) 721–726.
[29] K.A. Mehl, J.M. Davis, J.M. Clements, F.G. Berger, M.M. Pena, J.A. Carson, Decreased intestinal polyp multiplicity is related to exercise mode and gender in ApcMin/+ mice, J. Appl. Physiol. 98 (1985) (2005) 2219–2225.
[30] A. Nawaz, A. Aminuddin, T. Kado, A. Takikawa, S. Yamamoto, K. Tsuneyama, et al., CD206(+) M2-like macrophages regulate systemic glucose metabolism by inhibiting proliferation of adipocyte progenitors, Nat. Commun. 8 (2017) 286.
[31] A. Palazon, P.A. Tyrakis, D. Macias, P. Velica, H. Rundqvist, S. Fitzpatrick, et al., An HIF-1 alpha/VEGF-A Axis in cytotoxic T cells regulates tumor progression, Cancer Cell 32 (2017) 669–683.
[32] C.A. Bernhard, C. Ried, S. Kochanek, T. Brocker, CD169+ macrophages are sufficient for priming of CTLs with specificities left out by cross-priming dendritic cells, Proc. Natl. Acad. Sci. U. S. A. 112 (2015) 5461–5466.
[33] T. Haas, S. Heidegger, A. Wintges, M. Bscheider, S. Bek, J.C. Fischer, et al., Card9 controls Dectin-1-induced T-cell cytotoxicity and tumor growth in mice, Eur. J. Immunol. 47 (2017) 872–879.
[34] L.E. Kropp, M. Garg, R.J. Binder, Ovalbumin-derived precursor peptides are transferred sequentially from gp96 and calreticulin to MHC class I in the endoplasmic reticulum, J. Immunol. 184 (2010) 5619–5627.
[35] S.E. Hardison, G.D. Brown, C-type lectin receptors orchestrate antifungal immunity, Nat. Immunol. 13 (2012) 817–822.
[36] S. Chiba, H. Ikushima, H. Ueki, H. Yanai, Y. Kimura, S. Hangai, et al., Recognition of tumor cells by Dectin-1 orchestrates innate immune cells for anti-tumor responses, Elife 3 (2014) e04177.
[37] J. Yang, L. Chai, C. Gao, T.C. Fowles, Z. Alipio, H. Dang, et al., SALL4 is a key regulator of survival and apoptosis in human leukemic cells, Blood 112 (2008) 805–813.
[38] M. Canestraro, S. Galimberti, H. Savli, G.A. Palumbo, D. Tibullo, B. Nagy, et al., Synergistic antiproliferative effect of arsenic trioxide combined with bortezomib in HL60 cell line and primary blasts from patients affected by myeloproliferative disorders, Cancer Genet. Cytogenet. 199 (2010) 110–120.
[39] S. Ganesan, A.A. Alex, E. Chendamarai, N. Balasundaram, H.K. Palani, S. David, et al., Rationale and efficacy of proteasome inhibitor combined with arsenic trioxide in the treatment of acute promyelocytic leukemia, Leukemia 30 (2016) 2169–2178.
[40] W. Blum, S. Schwind, S.S. Tarighat, S. Geyer, A.K. Eisfeld, S. Whitman, et al., Clinical and pharmacodynamic activity of bortezomib and decitabine in acute myeloid leukemia, Blood 119 (2012) 6025–6031.
[41] H. Yang, Y.A. Minamishima, Q. Yan, S. Schlisio, B.L. Ebert, X. Zhang, et al., pVHL acts as an adaptor to promote the inhibitory phosphorylation of the NF-kappaB agonist Card9 by CK2, Mol. Cell 28 (2007) 15–27.
[42] J. Liu, L. Gunn, R. Hansen, J. Yan, Combined yeast-derived beta-glucan with antitumor monoclonal antibody for cancer immunotherapy, Exp. Mol. Pathol. 86 (2009) 208–214.
[43] M. Liu, F. Luo, C. Ding, S. Albeituni, X. Hu, Y. Ma, et al., Dectin-1 activation by a natural product beta-glucan converts immunosuppressive macrophages into an M1-like phenotype, J. Immunol. 195 (2015) 5055–5065.