Mini-review
Galectin-3 – A jack-of-all-trades in cancer
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Abstract
Galectin-3 is a mammalian β-galactoside-binding protein that is expressed by various types of human cells. Changes in galectin-3 expression and subcellular and intercellular localizations are commonly seen in cancer and pre-cancerous conditions. It is increasingly recognized that galectin-3 is an important regulator of a broad range of cancer cell activities and plays important roles in cancer cell growth, transformation, apoptosis, angiogenesis, adhesion, invasion and metastasis. Such a divergent influence of galectin-3 on cancer cell activities derives from its multiple inter- and sub-cellular localizations where it interacts with a range of different binding partners. This mini-review summarises the diverse influences of galectin-3 on cancer cell behaviours with particular emphasis on its role in tumorigenesis and metastasis.

1. Introduction
Galectins are a family of mammalian β-galactoside-binding proteins that share highly conserved carbohydrate recognition domains (CRDs) [1]. To date, 15 galectin members have been identified and they are classified into three subgroups depending on their structural differences and the number of CRDs within their polypeptide chains [2]. The prototypical galectins (galectin-1, -2, -5, -7, -10, -11, -13, -14 and -15) contain one CRD whereas the tandem-repeat galectins (galectin-4, -6, -8, -9 and -12) have two CRDs that are separated by a linker region in a single polypeptide chain. Galectin-3 is the exclusive member of the chimera-type galectin subgroup and contains one CRD that is connected to an extended non-lectin N-terminal domain (Fig 1).

Galectin-3 comprises of one CRD at its C-terminus and an unusual flexible 110–130 amino acids, N-terminal domain (ND) [4,5]. The galectin-3 ND consists of 7–14 repeats of a 9-amino acid sequence: Pro-Gly-Ala-Tyr-Pro-Gly-X-X-X, and is sometimes referred to as a collagen-like ND due to its homology to collagen α1 (II) chain [6]. The structure of galectin-3 remains highly conserved across animal kingdom and is also unique in the galectin family [7].

The ND of galectin-3 is essential for galectin-3 multimerization and is sensitive to proteolysis by MMP-2 and MMP-9 matrix metalloproteinases [8,9]. The first 12 amino acids of the galectin-3 ND are necessary for galectin-3 secretion as well as for galectin-3 nuclear translocation [10,11] whilst the highly conserved Ser6 residue is involved in the anti-apoptotic activity of galectin-3 [12]. It has been reported that the Tyr102 residue at the ND may differ from laboratories [4]. Changes in galectin-3 expression are commonly seen in cancer and pre-cancerous conditions [2]. Increasing evidence shows that galectin-3 is involved in the regulation of diverse cancer cell activities that contribute to tumourigenesis, cancer progression and metastasis.

1.1. Galectin-3 structure

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also participate in galectin-3 carbohydrate binding activity [13,14].

The C-terminal CRD of galectin-3 contains approximately 130 amino acids that form a globular structure with five- and six-stranded \(\beta\)-sheets arranged in a \(\beta\)-sandwich [13]. The CRD contains a NWGR (Asp-Trp-Gly-Arg) motif which is essential for galectin-3 binding to \(\beta\)-galactosides [15,16]. This NWGR motif is also present in the B-cell lymphoma 2 (Bcl2) family members of apoptosis regulators and is responsible for the anti-apoptotic activity of galectin-3 [17].

Galectin-3 binds more strongly to galactose-terminated glycans than to simple galactose. For example, lactose (Lac) and N-acetylactosamine (LacNAc) are much stronger ligands for galectin-3 than galactose. LacNAc also shows to have a 5-times higher affinity for galectin-3 than Lac [13,18,19]. The galectin-3 ND has been shown to also contribute to the increased binding affinity of galectin-3 to larger oligosaccharides (extended Lac or LacNAc) as compared to other galectins [20]. This is due to a conformational change of galectin-3 after its initial interaction with its carbohydrate ligands [18]. Galectin-3 can also interact with many intracellular proteins by protein–protein interactions in a carbohydrate-independent manner [21–25].

1.2. Galectin-3 localisation and multimerisation

Galectin-3, synthesized in the cell cytoplasm, shuttles between the cytoplasm and nucleus. It can also be secreted to the cell surface and biological fluids [4] through a non-classical exocytosis process [11,26]. Galectin-3 translocation from cytoplasm to the nucleus is attributed to its ND [10] although some evidence also suggests the involvement of its CRD [27]. The movement of galectin-3 from nucleus to the cytoplasm, on the other hand, involves a nuclear export sequence located within its CRD [28]. In the cytoplasm, galectin-3 can bind to Bcl-2 and inhibit cell apoptosis [17]. Cytoplasmic galectin-3 can also interact with the activated GTP-bound K-Ras [21,29] and affect Ras-mediated Akt signaling [30,31]. In the nucleus, galectin-3 promotes pre-mRNA splicing and participates in spliceosome assembly via complexes with nuclear protein Gemin4 [2,4]. Nuclear galectin-3 can also regulate gene transcription by enhancing transcription factor association with Spi1 and CRE elements in the gene promoter sequences [4].

In solution, galectin-3 largely occurs as a monomer [32]. It can also form homodimer by self-association through its CRDs in the absence of its binding ligands [33]. However, in the presence of its carbohydrate binding ligands, galectin-3 can polymerize up to pentamers through its ND (Fig. 1) [8,33,34]. Multimerisation is a common feature of extracellular galectin-3, where it often cross-links its cell surface ligands to form lattice-like structures [34,35] and triggers the initiation of cell surface molecule-associated cell signaling [2,36,37].

Galectin-3 is widely expressed in human tissues, including immune cells, epithelial cells and sensory neurons [4,38]. During early stages of human embryogenesis, however, galectin-3 shows a more specific expression pattern, mainly in the epidermis, kidney, chondrocytes and the liver [4]. Interestingly, galectin-3 knock-out mice remain viable and display no obvious abnormalities [16], indicating possible redundant or replaceable role of galectin-3 by other galectin members in normal physiological conditions. Conversely, galectin-3 overexpression is a predominant feature of many cancers. There is also a general shift of galectin-3 localization from nucleus to the cytoplasm in cancer development from adenoma to carcinoma [39,40]. There is strong evidence showing the involvement of altered galectin-3 expression and localization in the regulation of a broad range of cancer cell activates,
such as cancer cell growth, transformation, apoptosis, immunosuppression, angiogenesis, adhesion, invasion and metastasis (Fig 2).

1.3. Galectin-3 in tumour transformation

Substantial evidence indicates that galectin-3 overexpression promotes neoplastic transformation [21,36,41,42]. In highly malignant human breast carcinoma cells, suppression of galectin-3 resulted in reversion of the altered cell morphology as well as in loss of anchorage- and serum-independent growth and led to the inhibition of tumour growth in immunologically suppressed mice [41]. Likewise, inhibition of galectin-3 expression by antisense cDNA in human thyroid papillary carcinoma cells led to suppression of anchorage-independent cell growth [42]. Furthermore, transfection of galectin-3 cDNA into normal thyroid follicular cells resulted in altered gene expression, loss of contact inhibition and serum-independent growth [43]. These results suggest that the maintenance of transformed phenotype of malignant breast and thyroid cells is closely associated with galectin-3 expression.

The molecular mechanism of galectin-3-mediated transformation has shown to be partly attributed to the galectin-3 interaction with oncogenic K-Ras [21]. The galectin-3-Ras interaction enhances the Ras anchorage to the plasma membrane that causes constitutive activation of Ras-dependent phosphatidylinositol 3-kinase (PI3-K) and Raf-1 activation. The tumourigenic potential of galectin-3 may also involve the interactions of galectin-3 with β-catenin or various transcription factors that enhance the expressions of cyclin D and c-MYC [2,25] and promote cell cycle progression [4].

1.4. Galectin-3 in regulation of apoptosis

Cytoplasmic galectin-3 is a well-known apoptotic inhibitor. Following apoptotic stimuli, cytoplasmic galectin-3 interacts with a calcium-dependent and phospholipid-binding protein synxin. This galectin-3-synxin interaction helps the translocation of galectin-3 to the mitochondria [44] where it interacts with Bcl-2 and blocks the alteration of mitochondrial membrane potential and cytochrome c release [44,45]. Nuclear galectin-3, on the other hand, has been shown to promote cell apoptosis in human prostate cancer cells [46,36]. The mechanism of this pro-apoptotic action of galectin-3 remains largely unknown but has been suggested to be linked with galectin-3 interaction with an apoptosis-associated protein Nucling [47]. Thus, galectin-3 can be both anti- and pro-apoptotic dependent largely on its sub-cellular localizations.

1.5. Galectin-3 in immune regulation

Extracellular galectin-3 can bind to the cell surface N-glycans and induce monocyte and T cell apoptosis during tumour cell evasion from the immune surveillance. Galectin-3 can suppress IL5 production and inhibit B lymphocyte differentiation. There is also evidence that galectin-3 can increase phagocytosis of neutrophils [48] and oxidative response of immune cells [49]. In human monocytes, galectin-3 is chemotactic and increases Ca²⁺ influx at high concentration (1 μM), whereas at low concentration (10–100 nM) it promotes chemokinesis [4,50]. Thus, galectin-3 regulates immune cell activities and contributes to immunosuppression in tumorigenesis and metastasis.

**Fig. 2.** Divergent effects of galectin-3 on cancer cell activities at various intercellular and extracellular localizations.
1.6. Galectin-3 in angiogenesis

Formation of new blood vessels from pre-existing vessels – angiogenesis – is a crucial step in tumour cell invasion and metastasis [36]. Angiogenesis fuels tumours with essential oxygen and nutrients for excessive tumour growth. Exogenous galectin-3 has shown to enhance endothelial cell motility in vitro in cell culture and promotes new capillaries formation in vivo in mice [51]. This angiogenesis promoting effect of galectin-3 involves the interaction of galectin-3 with integrins or glycans expressed on cell surface molecules [52]. This causes clustering of galectin-3 with its cell surface ligands and the activation of focal adhesion kinase that leads to modulation of vascular endothelial growth factor (VEGF)- and basic fibroblast growth factor (bFGF)-mediated angiogenesis [53]. An interaction of galectin-3 with the endothelial cell surface enzyme aminopeptidase N/CD13 has also been reported to regulate endothelial vascularisation in the early steps of angiogenesis [54].

1.7. Galectin-3 in cancer cell adhesion

Tumour cell adhesion to and subsequent invasion through the extracellular matrix (ECM) at primary tumour sites are essential steps in epithelial cancer metastasis. Galectin-3 overexpression is associated with an increased invasiveness of many types of tumour cells such as neuroendocrine tumour pheochromocytoma, ovarian, melanoma, thyroid and colorectal cancer cells [4,55]. Overexpression of galectin-3 enhances tumour cell adhesion to ECM and promotes the escape of tumour cells from the primary tumour sites [37]. Such an effect of galectin-3 is attributed partly by its interaction with a range of the ECM glycoproteins such as fibronectin, collagen IV, elastin, and laminin [4]. The interaction of galectin-3 with cancer cell surface glycans such as the Mga5-modified N-glycans [56], laminin and lysosome-associated membrane glycoproteins [4,55] has also shown to be involved in galectin-3-mediated cancer cell adhesion. The interaction of galectin-3 with cell surface epidermal growth factor receptor (EGFR) and transforming growth factor-β receptor (TGFβR) is believed to also contribute to the increased invasiveness of tumour cells. In this case, galectin-3 crosslinks these cell surface molecules and prevent their endocytosis and signalling.

1.8. Galectin-3 in tumour cell metastatic spreading

Survival of the invaded tumour cells in the blood/lymphatic circulation and their subsequent adhesion to the blood vascular endothelium comprises of important steps in cancer metastasis. Recent investigations have revealed that circulating galectin-3 may play a very important role in these processes [57]. Compared with that in healthy people, the level of circulating galectin-3 is significantly higher in the bloodstream of cancer patients in particular those with metastasis [58]. Higher level of circulating galectin-3 is associated with increased metastasis and poor prognosis in several types of cancers including colorectal, thyroid and ovarian cancer. It has been demonstrated recently that galectin-3 binds to the oncofetal Thomsen-Friedenreich (galactoseβ1,3 N-acetylgalactosamine, TF) antigen on the transmembrane mucin MUC1 expressed on tumour cells [59]. The binding of galectin-3 to cancer-associated TF/MUC1 induces redistribution of MUC1 on the cell surface that allows the exposure of the smaller cell surface adhesion molecules which otherwise are concealed by the large and heavily glycosylated cell surface MUC1. This results in increased cancer cell heterotypic adhesion to the blood vascular endothelium [60] as well as increased cancer cell homotypic aggregation for the formation of tumour micro-emboli [61]. The formation of tumour microemboli enhances the survival of disseminating tumour cells in the circulation by preventing initiation of anoikis. Moreover, galectin-3 expressed on cancer as well as on endothelial cell surface can itself act as a cell adhesion molecule by interaction with the cancer- and endothelial-associated TF disaccharides and increase cancer cell heterotypic adhesion to vascular endothelium and cancer cell homotypic aggregation in cancer cell haematogenous dissemination in metastasis [62–64].

2. Conclusion remarks

As a multifunctional protein widely expressed by many types of human cells, galectin-3 overexpression and changes of sub- and inter-cellular localization are commonly seen in various types of human cancers. Although much remains to be further explored, more and more evidence indicates that galectin-3 is involved in the regulations of a myriad of cancer cell activities during cancer development, progression and metastasis. This is hugely attributed to the altered interactions of galectin-3 with a range of different binding partners at different locations in cancer conditions. Targeting the actions of galectin-3 will represent a promising therapeutic strategy for the development of effective therapeutic agents for cancer treatment.

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References


