




The role of Galectin-3 in modulating tumor growth and immunosuppression within the tumor microenvironment

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ABSTRACT

The efficacy of cancer immunotherapy is limited, in part, by the multitude of immunosuppressive mechanisms present within the tumor microenvironment (TME). Galectin-3 (Gal-3) is a lectin that contributes to TME immunosuppression and regulates diverse functions including cellular homeostasis and cancer biology. Increased Gal-3 expression during cancer progression augments tumor growth, invasiveness, metastatic potential, and immune suppression, which highlights the potential use of Gal-3 as a therapeutic target capable of modulating anti-tumor immunity. Here, we discuss the mechanisms by which Gal-3 regulates lymphocytes, the role of Gal-3 in lung and prostate tumors, and the contribution of Gal-3 to TME immunosuppression.

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Introduction

The immune system is designed to protect the host against disease, and possesses numerous mechanisms that tightly regulate its activity to prevent autoimmune reactions against normal, healthy host cells. However, these same regulatory mechanisms may be hijacked by cancerous cells, thereby allowing them to escape detection and attack by the immune system. Specifically, the tumor microenvironment (TME), which represents a complex ecosystem involving innumerable interactions between immune cells, cancer cells, stromal cells, and the extracellular matrix, can support tumor proliferation, survival, and metastasis and is highly immunosuppressive.¹⁻³ The TME achieves immunosuppression through a myriad of different ways; for example, tumor-associated macrophages, cancer-associated fibroblasts, and tumor cells can all secrete suppressive cytokines and chemokines, and there can be metabolic competition over consumption of nutrients by tumor cells, or a shortage of oxygen. Other immunosuppressive mechanisms include the production of inhibitory metabolites, migration failure due to rigid extracellular matrix, poor antigen presentation, chronic T cell receptor (TCR) signaling, and inhibitory receptor expression by tumor cells and stromal cells.⁴ An additional important regulatory mechanism at play in the TME occurs through the glycoprotein Galectin-3 (Gal-3). Gal-3 binds the TCR in the immunological synapse on the cell surface, thereby restricting TCR movement, potentiating TCR downregulation, and suppressing early activation of T cells through the TCR signaling pathway.^{5,6}

Gal-3 is a structurally unique glycoprotein that has been studied extensively in different disease contexts including

fibrosis, inflammation, and cancer. Gal-3 is a member of the lectin family, of which 14 mammalian galectins have been identified. Mammalian galectins have binding specificity to β -glycoside structures and are classified into three groups based on their conserved carbohydrate-recognition-binding domain (CRDs) structures: prototypes, tandem repeat, and chimera groups.⁷ Galectin-1, -2, -5, -7, -10, -11, -13, and -14 are members of the prototype galectin group that contain only one CRD. Members of the tandem repeat group (galectin-4, -6, -8, -9, and -12) contain two distinct CRDs connected by a non-conserved 70 amino acid linker sequence that enables each galectin to bind two carbohydrate epitopes. The chimera galectin group contains only one member, Gal-3, which contains one CRD like the prototype group, but the CRD in Gal-3 is connected to a unique N-terminal domain of about 120 amino acids that are rich in proline and glycine.⁸ Gal-3, with only one CRD, can form homo-dimers and oligomers through its N-terminal domain depending on the concentration and availability of the ligands. Further, the oligomeric structure of Gal-3 contributes to its function,⁹ as the oligomeric form of Gal-3 allows Gal-3 to perform biological functions not performed by all other galectins.^{10,11} The oligomeric forms of Gal-3 only form in the extracellular space (Fig. 1), where Gal-3 oligomers can bind substrates through its CRD domain and induce intracellular signal transduction through clustering surface proteins, cell-cell interactions, or cell to extracellular matrix (Fig. 1).¹² Furthermore, Gal-3 function depends not only on its oligomeric state in the extracellular space but also on its subcellular location, where Gal-3 monomers can be found either in the cytoplasm or nucleus (see ref. 9 for additional details of the subcellular

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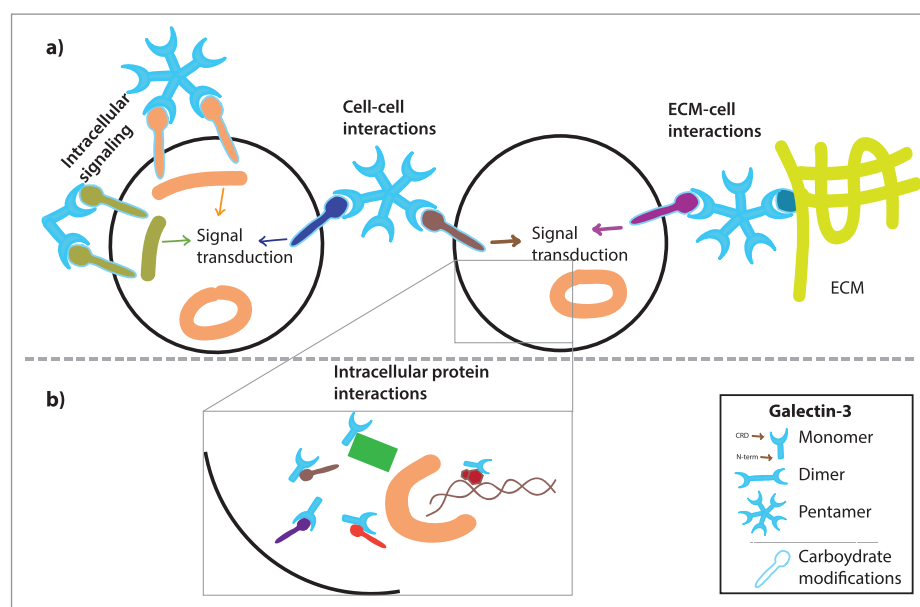


Figure 1. Gal-3 binds substrates through different mechanisms depending on its cellular location. a) In the extracellular space, Gal-3 binds to its substrate through CRD-specific carbohydrate modifications on the substrate. Gal-3 binding to these substrates can induce surface protein interactions, cell-cell and cell to extracellular matrix interactions, which can all lead to initiation of downstream signaling. b) In the intracellular space, Gal-3 binds to its substrates through direct protein-protein interactions between Gal-3 and the substrate.

location of Gal-3 and its function). There is no known difference in the generation of extracellular versus intracellular Gal-3. The signal that determines the subcellular localization and the mechanism of extracellular Gal-3 secretion are not clear. In the intracellular space, Gal-3 binds substrates through protein-protein interactions, for example, intracellular Gal-3 binds Bcl-2 to inhibit apoptosis.^{9,13,14}

On a cellular level, Gal-3 expression is dynamic during mouse development. The earliest Gal-3 protein is detected in the cells of trophectoderm of blastocyst,¹⁵ and then a few days later in gestation Gal-3 is exclusively found in the notochord cells.¹⁶ In later stages of development, Gal-3 protein is found in the cartilage of vertebrae, ribs and facial bones, the suprabasal layer of epidermis, the endodermal lining of the bladder, larynx, and esophagus. The liver and lungs, as well as the mineralizing part of the bones and some other organs, have a punctate Gal-3 distribution that is associated with macrophages and/or related cell types such as osteoclasts.¹⁵ In adult mice, Gal-3 is mainly restricted to epithelial and myeloid cells. Within these cell types, Gal-3 can be either localized in the nucleus, cytoplasm, plasma membrane or secreted into extracellular space, however the primary location is in the cytoplasm.

The cellular presence of Gal-3 in humans is similar to what is observed in mice. During the first trimester of human embryogenesis, Gal-3 is found in epithelia such as the skin, epithelial lining of the digestive and respiratory tract, urothelium and excretory tubes of the kidney, myocardial cells, peripheral and pre-ossifying hypertrophic chondrocytes, as well as in the notochord and liver.¹⁷ Notably, Gal-3 protein is not found in lymphoid tissues and naïve lymphocytes do not express Gal-3; however, sufficient stimuli (both mitogen and IL-2) can induce lymphocytes to produce intracellular Gal-3, which is not secreted into the extracellular space.¹⁸ Gal-3 protein is also not found in cells of the central nervous system. According to the Human Protein Atlas project, in adults, the majority of

epithelial cells and variety of normal tissue types contain both cytoplasmic and nuclear Gal-3 protein (Fig. 2).¹⁹ The same study showed little or no Gal-3 in hepatocytes, neuronal cells, and most lymphoid cells along with microglia and astrocytes.¹⁹

The presence of Gal-3 across most adult tissue types illustrates the necessity of Gal-3 in basic biology. However, increased levels of Gal-3 in the tumor and TME in several cancer types suggests a contribution to immunosuppression and a role in promoting tumor growth. Given the critical role the TME plays in regulating tumor progression and metastasis, recent studies have focused on new therapeutic strategies that will turn the TME from an immunosuppressive to immunostimulatory. Cancer immunotherapies such as checkpoint blockade, T cell agonists, and adoptive cell therapy have been successful at producing effective antitumor responses within the TME. However, these immunotherapies are often not sufficient to overcome the complex immunosuppressive nature of the TME. Given this impediment in improving cancer immunotherapy, understanding the glycoprotein interactions within the TME, such as the immunosuppressive effects of Gal-3, will help us to overcome the challenges of immunosuppression. This review focuses on how Gal-3 affects immune cells, how Gal-3 contributes to immunosuppression in lung and prostate tumors (tumor types selected due to the differential expression of Gal-3 in these tumors compared to other solid tumors) and within the TME, and how targeting Gal-3 has evolved in cancer immunotherapy.

Galectin-3 in immune cells

Gal-3 is known to influence immune cells and can negatively regulate their function in pathological settings. Here, we focus on the effects of Gal-3 specifically on lymphocytes and macrophages because of their critical contribution to anti-tumor responses in the TME.

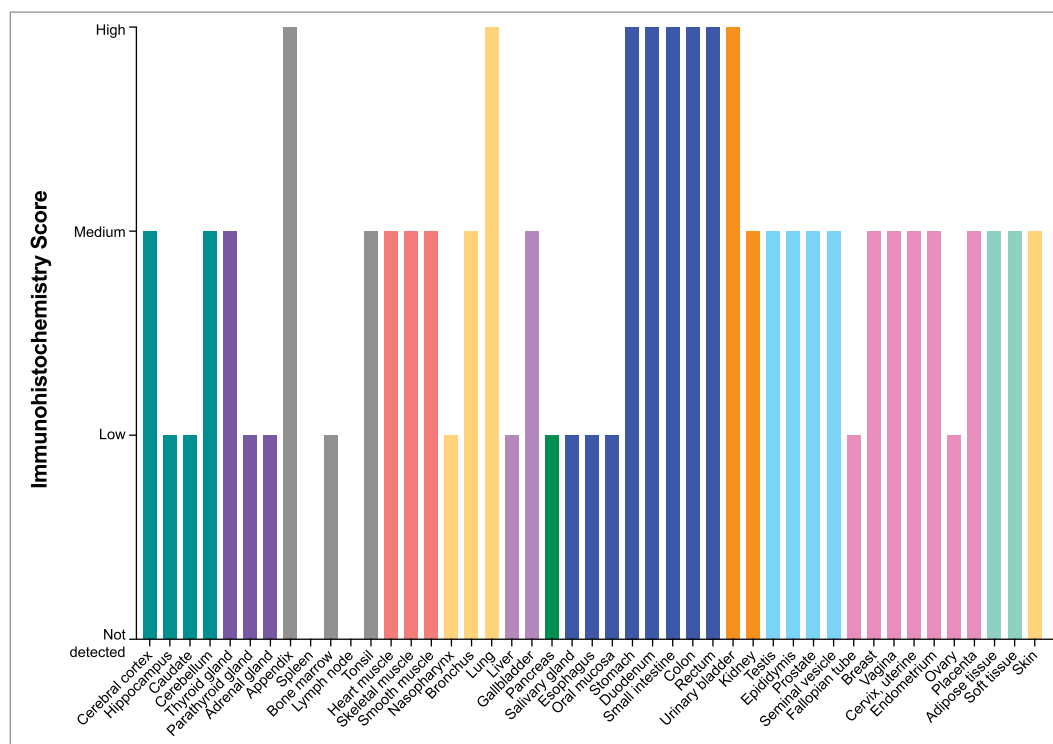


Figure 2. Gal-3 protein expression levels in different human tissue. Each color represents an organ system and its sub-tissues. All images of immunohistochemistry are manually annotated considering staining intensity, fraction of stained cells, and subcellular localization using fixed guidelines for classification (adapted from The Human Protein Atlas).¹⁹ Within these tissue types, Gal-3 can be localized in the nucleus, cytoplasm, plasma membrane or secreted into the extracellular space, however the primary location is in the cytoplasm.

Gal-3 effects on lymphocytes

Due to the expression of Gal-3 in normal tissue and its participation in a vast range of functions, it has been challenging to investigate the role of Gal-3 in lymphocytes *in vivo*; however, for decades, researchers have studied the role of Gal-3 in mediating lymphocyte suppression *in vitro*. Further extracellular and intracellular Gal-3 can have disparate effects on lymphocyte function, which complicates the understanding of Gal-3 function. Extracellular Gal-3 secreted by tumor or normal cells regulates several important lymphocyte functions such as apoptosis, activation, TCR signaling, migration, adhesion, and IL-5 production. Notably, the cellular location of Gal-3 determines whether it has apoptotic or anti-apoptotic effects on T cells—extracellular Gal-3 induces apoptosis, while intracellular Gal-3 inhibits apoptosis. Extracellular Gal-3 induces apoptosis in human thymocytes and T cells by directly binding the glycoprotein receptors CD45 and CD71.²⁰ In contrast, overexpression of Gal-3 within the intracellular compartment of Jurkat T cells inhibited apoptosis induced by anti-Fas antibody and staurosporine.¹³ Intracellular Gal-3 is also involved in promoting cell growth and enhancing TCR signaling.^{13,21} Furthermore, Gal-3 knockout CD4 T cells exhibited increased TCR expression and higher IFN- γ secretion compared to wild-type CD4 T cells.⁵

The role of Gal-3 specifically within CD8 T cells is a topic of intense interest due to the vital role CD8 T cells play in immunotherapy. Despite this, little is known about the effects of either extracellular or intracellular Gal-3 on CD8 T cell function. In one experiment, extracellular Gal-3 had a suppressive effect on CD8 T cells as a Gal-3-deficient melanoma tumor cell

line or its supernatant cultured with tumor-reactive CD8 T cells induced a significant expansion and increase in IFN- γ levels in the CD8 T cells compared to co-cultures with Gal-3-expressing tumor cell lines or supernatant.^{22,23} In human tumor-derived CD8 T cells, Gal-3 expression has been associated with the loss of TCR and CD8 marker localization at the immunological synapse and subsequent loss of effector function.^{24,25} A recent study showed that extracellular Gal-3 binds to lymphocyte activation gene 3 (LAG-3) on CD8 T cells and possibly suppresses CD8 T cell function.²⁶ However, in most studies the effect of Gal-3 on CD8 T cells was shown in total splenocytes and/or in the presence of CD4 T cells, thus these data may not reflect direct effects of Gal-3 on CD8 T cells.

Galectin-3 effects on macrophages

Macrophages play an important role in host defense and maintenance of tissue homeostasis. Macrophages are a functionally heterogeneous cell population and depending upon the micro-environmental stimuli they can polarize into two main groups, M1 and M2. M1 are classically activated macrophages whose activating stimuli are interferon- γ (IFN- γ) and lipopolysaccharide (LPS). M2 are alternatively activated immunosuppressive macrophages, which include the subtypes M2 a (exposure to IL-4 or IL-13) and M2 c (exposure to IL-10 or glucocorticoids).²⁷ Gal-3 is highly expressed and secreted by macrophages themselves, which suggests a role for Gal-3 in the innate physiology of these cells.²⁸⁻³⁰

Classical M1 macrophage activation with LPS inhibits Gal-3 expression and release, whereas alternative

macrophage activation by IL-4/IL-13 leads to the accelerated biosynthesis and secretion of Gal-3,^{31,32} suggesting that Gal-3 may be a specific and highly up-regulated marker of M2-type macrophages.³³ IL-4 mediates M2 macrophage activation and subsequently activates increased Gal-3 expression as well as other phenotypic M2 activation markers. Gal-3 then becomes part of a feedback loop for driving M2 macrophage activation by binding to CD98 or CD98 and β 1 integrin complex, which leads to PI3 K activation and thus M2 activation. This IL-4-driven M2 macrophage activation is blocked by an extracellular Gal-3 carbohydrate binding inhibitor, bis-(3-deoxy-3-(3-methoxybenz-amido)-D-galactopyranosyl) sulfane, and is also inhibited by the deletion of Gal-3, CD98, or inhibition of PI3 K using small interfering RNA (siRNA).³¹ Thus, Gal-3 can turn classical M1 macrophages into alternative M2 macrophages, a source of additional Gal-3 in the TME.

Galectin-3 contribution to immunosuppression

Several studies have reported that Gal-3 expression increases during cancer progression and this expression results in detrimental outcomes such as increased tumor progression, invasiveness, and metastatic potential.^{10,34-36} Interestingly, Gal-3 affects more cancer types than other galectins (Table 1).³⁵ The common function of Gal-3 in many different cancer types is reviewed elsewhere;^{9,10} this review will focus exclusively on Gal-3 expression in prostate and lung cancer due to their unique Gal-3 expression profile.

Galectin-3 expression and function in prostate cancer

In other cancer types, Gal-3 expression increases throughout disease progression,³⁷ whereas Gal-3 expression in prostate

cancer is notably different. Gal-3 is strongly expressed in the early stages of prostate cancer, but this expression gradually decreases over disease progression and is completely lost in advanced stage prostate cancer.³⁷⁻³⁹ Due to this gradient of expression over disease progression, Gal-3 expression (in prostate cancer vs. benign tissues) may be useful in predicting biochemical recurrence.^{38,40}

In prostate cancer, Gal-3 regulates two coordinated steps of the metastatic cascade: the metastatic cells adhering to the microvascular endothelium (heterotypic adhesion) and the metastatic cells aggregating through interactions of tumor-associated Thomsen-Friedenreich glycoantigen with Gal-3 (homotypic aggregation).³⁶ Knockdown of Gal-3 using siRNA in human prostate cancer PC3 cells reduces tumor growth, cell proliferation, cell migration, colony formation, and invasion.⁴¹ Another study using the human prostate cancer PC3 cell line showed that Gal-3 knockdown results in a perturbed cell-cycle progression, including cell-cycle arrest at the G1 phase, up-regulation of nuclear p21, and hypo-phosphorylation of the retinoblastoma tumor suppressor protein (pRb). Up-regulation of nuclear p21 and hypo-phosphorylation of pRb leads to cell cycle arrest, suggesting a regulatory role for Gal-3 in cell-cycle progression.³⁵ Gal-3 is also reported to be involved in osteoclastogenesis through binding myosin-2 A.⁴²

In addition to different oligomeric forms, Gal-3 can also take on a cleaved form after cleavage by matrix metalloproteinases (MMP)-2/-9. In mouse models of breast and prostate cancers this cleavage is associated with angiogenesis, tumor growth, and resistance to apoptosis.⁴³ Further, the levels of cleaved Gal-3 have been shown to increase with metastasis,⁴¹ suggesting that the loss of Gal-3 expression during disease progression in prostate cancer may reflect cleavage of Gal-3 on the cell surface. Once cleaved, the Gal-3 CRD binds with higher affinity to its carbohydrate ligand, but loses its ability to multimerize through its N-terminal domain, thus abrogating any Gal-3 biological function that depends on its dimer or pentamer formation.⁴⁴ For example, intact Gal-3 promotes osteoclastogenesis through localization with myosin-2 A, a suppressor of osteoclast differentiation. In contrast, the prostate bone metastases expressing cleaved Gal-3 can still bind to myosin-2 A, but only partially reduce osteoclast differentiation because cleaved Gal-3 cannot form multimers.⁴² Further investigation is necessary to elucidate the relationship between the loss of Gal-3 expression and Gal-3 cleavage in advanced prostate cancer.

Galectin-3 expression and function in lung cancer

The majority of studies investigating Gal-3 in lung cancer have suggested Gal-3 involvement in tumor initiation, metastasis, and migration.³⁴ Gal-3 expression varies among different types of lung cancer. For example, small cell lung cancer (SCLC) expresses Gal-3 at very low levels or not at all, while non-small cell lung cancer (NSCLC) expresses high levels of Gal-3.⁴⁵ Lung spheres of cancer stem cells derived from a NSCLC cell line (H1299) express relatively high levels of Gal-3 over serial passages compared to monolayer cells. Gal-3 knockdown in these NSCLC cell line-derived spheres decreased stemness-related genes, suggesting a co-factor role for Gal-3 by interacting with β -catenin to increase the transcriptional activity of downstream

Table 1. Summary of the effect of Galectin-3 on specific cancers^a.

Cancer Type	Gal-3 Expression	Consequences of Gal-3
Breast	increased expression	helps evade immune surveillance and killing of active T cells
Prostate	decreased expression over disease progression	regulates metastatic cascade
Cervical	increased expression	resistance to chemotherapy
Lung	increased expression in NSCLC	involved in regulating metastasis to lung
Gastric	increased expression	enhances gastric cell motility and mediates metastasis
Melanoma	increased expression	increased growth, progression, angiogenesis, and metastasis
Renal cell cancer	increased expression	anti-apoptotic, resistance to cytotoxic treatment
Bladder	increased expression	increases malignant potential
Pituitary	increased expression	cell proliferation and tumor progression
Thyroid	increased expression	increased progression of differentiated thyroid cancer
Pheochromocytoma	increased expression	predicts benign vs. malignant potential
Glioma	increased expression	activated in microglia and macrophages the glioma progresses

^aadopted from Ebrahim et al. Galectins in solid malignancies.

stemness-related genes. In addition, after Gal-3 knockdown the cell line lost its ability to initiate tumors and had decreased aggressiveness, clonogenicity, and chemoresistance to cisplatin or paclitaxel.⁴⁶ Furthermore, Gal-3 knockout resulted in attenuation of lung carcinogenesis. These experiments suggest a regulatory role for Gal-3 in carcinogenesis-related B-cell receptors, ERK/MAPK, and peroxisome proliferator-activated receptor (PPAR) signaling pathways. Furthermore, the 4-(methylnitrosamino)-1-(3-pyridyle)-1-butanone (NNK)-induced incidence of lung tumors were significantly lower in Gal-3 knockout mice compared to their Gal-3 positive counterparts.⁴⁷

Gal-3 is expressed on the endothelium of all the major compartments of the normal lung, and the presence of Gal-3 in normal lung tissue is thought to be a homing factor for lung metastasis because it provides a location for cells to adhere as they spread throughout the tissue.⁴⁸ For example, endothelium membrane-bound Gal-3 binds high affinity ligands poly-N-acetyllactosamine (polyLacNAc) on N-oligosaccharides on melanoma cells, thus providing adhesion for melanoma cells to metastasize into the lung. Manipulating Gal-3 expression, or expression of Gal-3 substrates in the lung after the onset of a primary tumor elsewhere, could be a means of controlling metastasis to lung. For example, down-regulation of the Gal-3 binding substrate polyLacNAc in melanoma cells resulted in a loss of adhesion, spreading, MMP-9 secretion, and motility of Gal-3 expressing cells. Due to the loss of these properties, melanoma cells also lost the ability to metastasize to the lung.⁴⁸ Because of the connection between Gal-3 expression and metastasis, Gal-3 has been explored for its use as a biomarker to predict metastasis in lung cancer, but the results have been mixed. Some studies demonstrated that high levels of Gal-3 and osteopontin in the serum and high expression of Gal-3 and osteopontin mRNA in NSCLC are associated with increased risk of developing metastasis and could be used as an index for evaluating undetectable NSCLC.⁴⁹ However, other

studies concluded that the expression and binding capacity of Gal-3 does not correlate with the staging of lung cancer and is therefore unfavorable as a prognostic marker.⁵⁰ Gal-3 involvement in metastasis to the lung makes Gal-3 a potential therapeutic target in order to eliminate homing of tumor metastasis into the lung.

Galectin-3 mediated immunosuppression in the TME

The TME consists of many different cell types including leukocytes, stroma, and neoplastic cells along with associated growth factors and chemokines. The functions of many of these TME components can be altered by the tumor to promote tumor proliferation and survival. For example, tumors promote immune suppression by inhibiting T cell activation, polarizing pro-tumor macrophages, and turning normal fibroblasts into cancer associated fibroblasts.⁵¹ Alteration of immune cells within the TME is one of the ways tumors escape immune control, which allows tumor progression and metastasis.

Gal-3 plays a crucial role in promoting tumor-driven immune suppression. In mixed lymphocyte cultures of T cells derived from peripheral blood mixed with autologous tumor cells, inhibition of tumor-expressed Gal-3 led to the expansion of high numbers of tumor-reactive T cells,²² suggesting that Gal-3 suppresses expansion of tumor-reactive T cells. Furthermore, Gal-3 secreted by tumor cells has been shown to alter macrophage polarization from M1 (anti-tumor macrophage) to M2 (pro-tumor macrophage), trigger CD8 T cell apoptosis, and restrict T cell receptor (TCR) clustering, all of which contribute to immunosuppression and facilitate tumor escape (Fig. 3).

Targeting Galectin-3 in immunotherapy

Due to the extensive role played by the TME in promoting tumor progression and metastasis, modulating the TME to

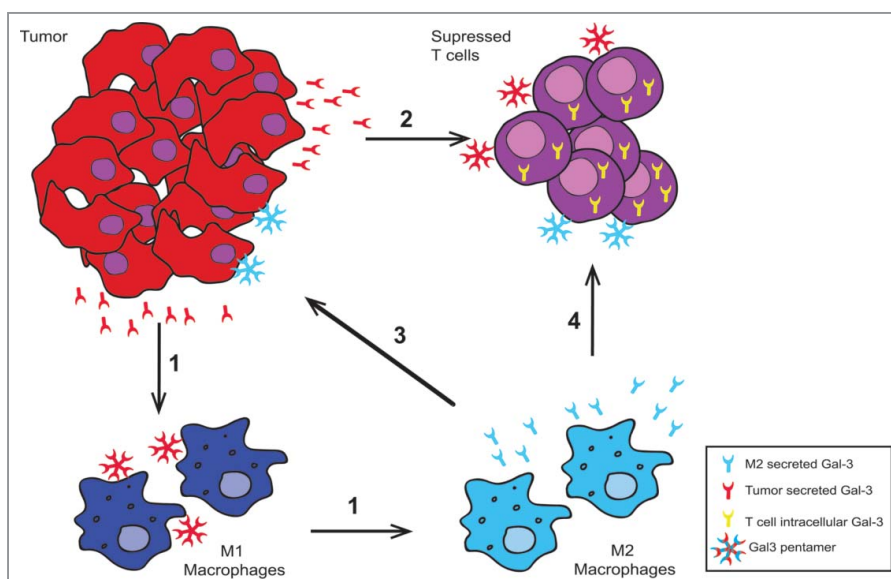


Figure 3. Impact of Gal-3 within the TME. Gal-3 is secreted by the tumor cells as monomers, which can form pentamers and bind substrates. The arrows indicate the influence of extracellular Gal-3 on various cell subsets. Gal-3 secreted by tumor cells: 1) polarizes M1 macrophages to M2 macrophages and 2) suppresses CD4 and/or CD8 T cells. Gal-3 secreted by M2 macrophages: 3) binds tumor cells to promote tumor progression/metastasis and 4) suppresses T cells.

decrease immunosuppression and increase immune activation has gained considerable attention in the field of cancer immunotherapy. The overwhelming evidence of Gal-3 involvement in boosting tumor growth, metastasis, and immune suppression has made Gal-3 an exciting target for cancer immunotherapy. The inhibition of Gal-3 in solid tumors in combination with T cell checkpoint blockade or T cell agonists has potential to augment anti-tumor immunity and improve tumor regression. In pre-clinical studies, our group has shown that treatment with a Gal-3 inhibitor, GR-MD-02, promotes antigen specific T cell expansion *in vivo*. In addition, GR-MD-02 combined with a stimulatory (agonist) anti-OX40 monoclonal antibody (mAb) improved survival in the MCA-205 sarcoma, 4T1 mammary carcinoma models, and TRAMP-C1 prostate cancer models.⁵² GR-MD-02 combined with anti-OX40 also reduced lung metastases in the 4T1 model.⁵² Furthermore, combination of GR-MD-02 with CTLA-4 or PD-1 checkpoint inhibitors has robust anti-tumor effects in multiple murine tumor models [Unpublished data]. This pre-clinical data provided the rationale for evaluating GR-MD-02 plus immunotherapy for patients with advanced cancer. GR-MD-02 was entered into two separate phase I clinical trials in combination with ipilimumab, a CTLA-4 inhibitor, or pembrolizumab, a PD-1 inhibitor, in patients with metastatic melanoma (anti-CTLA-4 or anti-PD-1), head and neck squamous cell carcinoma (anti-PD-1), and NSCLC (anti-PD-1) (NCT02575404; NCT02117362).

Another Gal-3 inhibitor, GCS-100, was used to treat elderly patients with relapsed chronic lymphocytic leukemia (CLL) in a phase II clinical trial, which resulted in partial responses in 6 out of 24 patients.⁵³ In a phase I clinical trial for treating resectable solid tumors, GCS-100 stabilized the disease in 16 of 24 patients.⁵⁴ Since the completion of these clinical trials, GCS-100 has not been used by any other cancer-specific clinical trials. However, recently GCS-100 was examined in preclinical mouse models. Mice bearing P815 tumors that were vaccinated and treated with GCS-100 exhibited 50% survival compared to controls treated with either GCS-100 or the vaccine, which all succumbed due to the tumors.²⁵ A recent preclinical study also showed that GCS-100 was efficacious in limiting disease progression and increasing survival of KRAS mutant NSCLC and pancreatic cancers and a KRAS-derived spontaneous cancer models.⁵⁵ In the light of this new pre-clinical data, GCS-100 may have a stronger anti-tumor response when used in combination therapy. These studies provide evidence for the potential efficacy of Gal-3 inhibition in combination with checkpoint blockade or T cell agonist immunotherapy for the treatment of cancer.

Conclusion

The uniquely structured lectin Gal-3 is expressed in most cell types in adults. The structure of Gal-3 allows it to oligomerize, which confers distinctive Gal-3 functions in situations of both homeostasis and pathological processes, such as cancer. Gal-3 expression typically increases during cancer progression, and this expression results in both enhanced suppression of the immune response and other detrimental outcomes including increased tumor progression, invasiveness, and metastatic potential. Recent data suggests that inhibiting Gal-3 in

combination with established immunotherapy has the potential to both alleviate immune suppression and decrease tumor growth. Given these promising preliminary results, additional studies are warranted to further investigate how Gal-3 contributes to tumor progression and the mechanisms by which Gal-3 inhibition combined with checkpoint blockade or T cell agonists augment cancer immunotherapy.

Disclosure of potential conflicts of interest

MF and ASR have no competing interests. WLR has received commercial research grants, consulting fees, and/or royalties from MedImmune, Bristol-Myers Squibb, Merck, Galectin Therapeutics, Nektar Therapeutics, Aeglea Biotherapeutics, IRX Therapeutics, Tesaro, Shire, and DNatrix.

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