REVIEW ARTICLE

Recent Advancements in Gremlin-1: Breast cancer

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Abstract

One of the bone's morphogenic protein (BMP) antagonists, Gremlin-1 or GREM-1, can bind directly to BMPs. GREM-1 can act in either BMP-dependent or -independent pathways, according to research. It reinforces organogenesis, tissue differentiation, and organ fibrosis. Recent research from numerous studies has demonstrated the significance of GREM-1 in the initiation, progression, and even metastasis of different cancers, including breast, cervical, gastric, and colorectal cancers. This review highlights the function of GREM-1 in the development of breast cancer and its effect on the cellular procedures and signalling pathways involved in carcinogenesis.

Keywords: Bone Morphogenetic, Carcinogenesis, Organogenesis, Colorectal Neoplasms, breast cancers, stem cells.

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Introduction

Breast cancer is the most frequently found cancer in women worldwide, with significant advancements in its diagnosis and treatment in past decades[1]. Breast cancer is among the types with a high risk of relapse and metastasis, but early detection responds positively to the treatment.[2]. Estrogen and estrogen receptors are commonly seen in the progression of breast cancer, which is treated by giving selective estrogen receptor modulators[3,4]. The most overexpressed receptor is the HER2 protein, which is the biomarker used in the treatment of HER2-positive breast cancer[3]. Despite breast cancer being the most common type of cancer worldwide, it still lacks research on targeted therapies for triple-negative cancers (lack of HER2, estrogen, and progesterone receptors)[5]. Unfortunately, identifying metastasis and recurrence-associated molecular markers of breast cancer remains obscure, thereby necessitating the need to identify therapeutic targets and mechanisms of action associated with breast cancer metastasis[6]

Bone morphogenetic proteins (BMPs) are tumour suppressor or oncogenes which plays a vital role in bone formation and other biological processes such as organogenesis and tissue differentiation[7]. Disadvantageously, the BMP antagonists bind directly to the BMP ligands and inhibit the BMP-induced signalling pathways[8].

Gremlin-1 or GREM1, a BMP antagonist, has a vital role in organ development, organ fibrosis , tissue differentiation and bone formation[9]. On the other hand, it is also involved in inflammation[10], and BMP-dependent or independent cancer[11,12]. GREM-1 is involved in breast cancer growth and is associated with a poor survival rate[11] (Figure 1). It is also found to be responsible for causing metastasis[13]. Epithelial-mesenchymal transition (EMT) is a phenomenon in which epithelial cells lose contact with each other and become mobile mesenchymal cells which promote cell migration and tumour growth under the influence of GREM-1[14]. It is

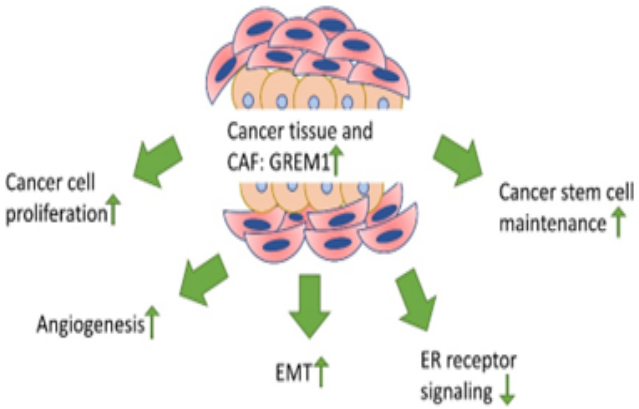

Figure-1: Role of Gremlin-1 in cancer tissue; Increased expression of Gremlin-1 in cancer cell tissue and CAFs (cancer-associated fibroblasts) results in angiogenesis, proliferation, epithelial to mesenchymal transition of cancer cells, and stem cell maintenance, hence, playing a major role in the growth and progression of cancer.

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found that GREM-1 also mimics the effects of vascular endothelial growth factor (VEGF) on the VEGF receptor-2 (VEGFR-2) in endothelial cells[15]. Recent clinical research has shown the correlation between GREM-1 and poor prognosis in various cancers, such as breast and colorectal cancer, where GREM-1 provides a route to the activation of PI3K/AKT/mTOR and antagonistic BMP2 signalling pathways[16]. Similarly, GREM-1 has a functional part in regulating EMT and the sensitivity of hepatoma cells to drugs like sorafenib in hepatocellular carcinoma[17].

In this review, we have highlighted GREM-1 and its correlation with breast cancer and stem cell maintenance via different pathways to better understand its mechanism of action.

Methods

The literature search was done on PubMed, Google Scholar, Scopus, and Embase. Mesh terms used for the search were "GREM-1", "Gremlin-1", "breast cancer" and "breast neoplasm. We screened 290 articles, published during 2010 to 2022 and identified 25 which had focused on the role of GREM-1 in breast cancer stem cells. The review focused on questions like the role of GREM-1 in different pathways, progression, and microenvironment of breast cancer stem cells, recent advancements, and GREM-1 as a marker for prognosis.

GREM-1 and BMP correlation

BMPs (Bone morphogenetic proteins), the subfamily of TGF- β (Transforming Growth Factor Beta), are extracellular proteins that play an important role in organogenesis and tissue differentiation[8]. BMPs act on cognate transmembrane serine/ threonine kinase receptors which result in the accumulation of activated R-SMADs (SMAD 1/5/8) (Receptor -Regulated Suppressor of mothers against decapentaplegic) that causes further transcriptional responses[18] (Figure 2). BMP antagonists such as Noggin[19], Coco[20], and GREM-1[21] disrupt BMP signalling in cancers like breast cancer [22]. GREM-1 inhibits BMP 2, BMP4, and BMP7[15]. After using In Situ hybridization (ISH), levels of GREM-1 were detectable in CAFs (fibroblast-like cells). In the study they were not detectable in cancer-adjacent normal tissues, adjacent cancer-free breast tissues, or epithelial cells of breast cancers[22]. , Intriguingly enough in a 2019 study, GREM-1 was found to be elevated in metastatic cancer cells even with increased co-expression of a BMP protein in tumour cells, exhibiting a direct link to decreased relapse-free survival (RFS) in ER-negative breast cancer cells (HR = 1.51 (1.2–1.9), p-value = 0.00037)[13, 21], ER-positive (HR = 1.19

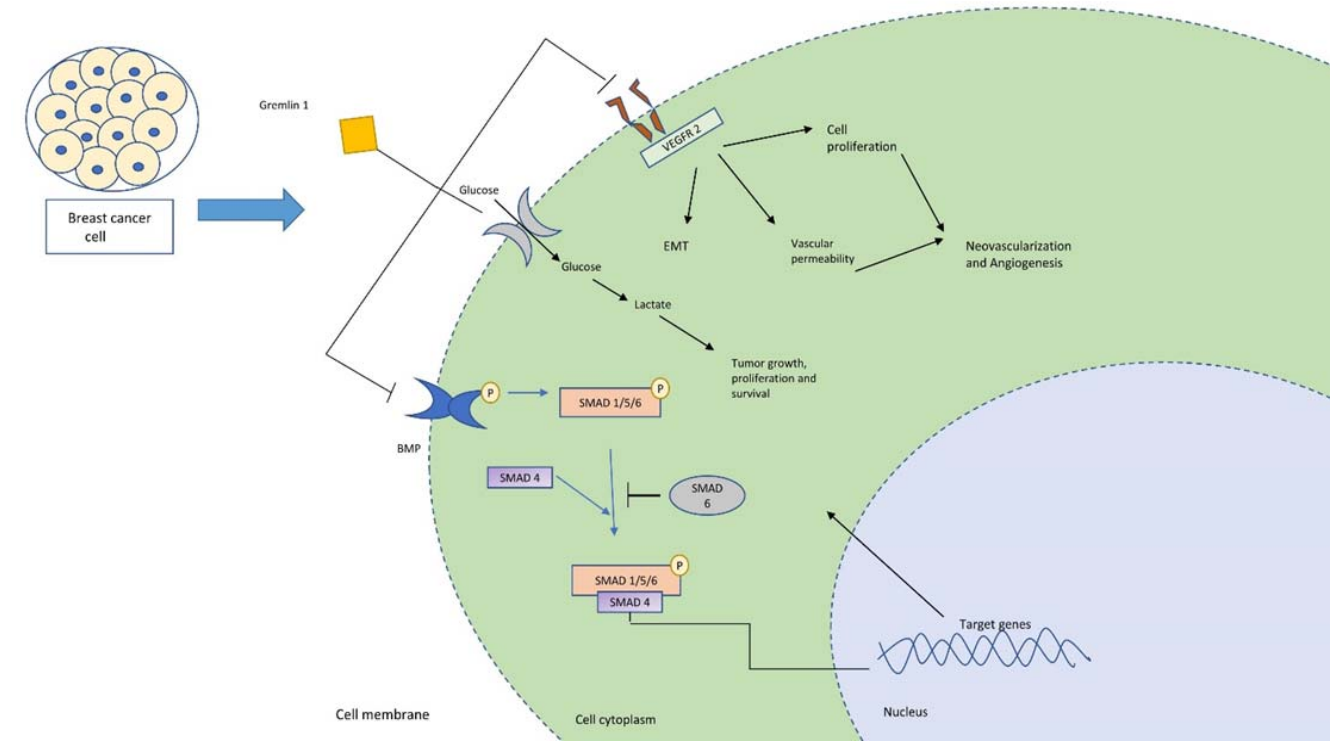

Figure-2: : Function of Gremlin-1; Gremlin-1 inhibits BMP and binds to BMP signaling pathway as well as vascular endothelial growth factor receptor (VEGFR) and also increases glucose uptake

(1.01–1.4), p-value = 0.035), human EGF receptor (HER)2+ and triple– (in all subtypes combined HR = 1.32 (1.18–1.47), p-value = 6.9e– 07), making GREM-1 a poor prognostic marker in all breast cancer subtypes[22]. Additionally, GREM-1 was one of the eight elevated genes found in invasive breast carcinoma patients compared to ductal carcinoma in situ patients[23], recommending that cancer cells form GREM-1 on their own and are of an invasive phenotype[13].

Role of GREM-1 in maintaining cancer cell stemness

Epithelial-origin cancer cells attain the undifferentiated form of stem cells by GREM-1 and also involved in the EMT process[22]. CD44+/high CD24–/low cells are known to stem population markers of breast cancer cells[24]. GREM-1 increases the expression of YAP, TAZ, SOX2, and OCT4 transcriptional regulators, thereby conserving breast cancer stemness and enhancing Mammo sphere formation[22]. GREM-1 generates epithelial to mesenchymal transition, a process where epithelial cells transition to mobile mesenchymal cells causing metastasis and cancer cell invasion[14] which aids in sustaining stem cell properties[25].

Many in vitro studies done in the past concluded that breast cancer stem cells are responsible for radiation and chemotherapy resistance[24]. The reason possibly is a decrease in pro-oxidants in CD44+/CD24– cells and the neoadjuvant trial design, which hypothesised that breast cancer stem cells are the reason for cytotoxic chemotherapy resistance[25]. After neoadjuvant chemotherapy, the number of atmospheres forming cells and CD44+/CD24– expressing cells increases[24].

GREM-1 and VEGFR-2 Angiogenesis is augmented by the VEGF which helps endothelial cellular proliferation, differentiation and enhancing microvascular permeability and vasodilation[26]. Sometimes GREM-1 mimics the role of VEGF and binds with VEGFR-2, which leads to a cascade of intracellular events which causes neovascularization and angiogenesis[27,28]. Thus, further research studies are required on the Gremlin-1/VEGFR2 axis as it is considered an excellent therapeutic target[29].

GREM-1 and Matrix Metalloproteinases (MMPs)

MMPs are endopeptidases whose level increases in most cancers, causing tumour proliferation, survival, angiogenesis, and metastasis by breaking down the ECM (extra-cellular matrix) and then finally modulating the tumour stroma[15]. There are many types of MMPS of which MMP13 is noted to be involved in the metastasis of breast cancer cells by Golgi membrane protein 1[15]. Correction analysis revealed a positive relation between GREM-1 and MMP13[21]. Sufficient literature is available indicating that the GREM-1/MMP13 axis hampered the breast cancer tumour[6,21,30,31]. GREM1 expression was compared with breast cancer and normal breast tissue using the Oncomine microarray database and other genes associated with angiogenesis, and metalloendopeptidase activity. The p-value calculated was 2.35 × 10–4 with a median rank of 27[21].

Role of GREM-1 in glycolysis of breast cancer cells

Grem 1 overexpressing breast cancer cells have been found to increase glucose uptake and lactate production via glycolysis[32], which leads to the lowering of the pH within the tumour microenvironment[33], considered to be a hallmark of cancer[34].

GREM-1 increases the expression of HK2[32]. Moreover, it stimulates the STAT3 transcription factor by the ROS-Akt signaling pathway[32]. Th.en ROS-Akt-STAT3 axis stimulates the GREM-1 which increases the activity of HK2 in breast cancer cells and there is increased uptake of glucose[32].

GREM-1 and ER-negative breast cancer

It has been shown that overexpression of GREM-1 in ER-negative breast cancer is linked to worse survival[11]. Few studies suggest that increased levels of GREM-1 had an association with oestrogen receptor expression[13]. It was found that all cell lines releasing GREM-1 lacked oestrogen receptor signalling[13]. In a recent study, overexpression of MMP 13 had a direct impact on reduced metastasis-free survival to a large extent in ER-negative breast cancer [21].

High mRNA levels of CRIM-1, GREM-1, and SMAD 6 (extracellular BMP antagonist) correlated with decreased relapse-free survival in ER-negative breast cancer[13]. In contrast, low mRNA levels of BMPER, CHRDL1, CRIM1, and SOSTDC1 correlated with reduced relapse-free survival in ER-positive breast cancer, suggesting that GREM-1 expression is linked to aggressive tumour progression.[13] In a study done by Park et al, GREM-1 mRNA was closely linked to RFS and distant metastasis-free survival (DMFS) in ER-negative breast cancer patients. ER- negative breast cancer patients were found to have high expression of Grem 1 supporting poor prognosis in this particular subtype (shown in table 1)[11] .

Conclusion and Future Direction

Research on GREM-1 is of great global importance since breast cancer is the most common cancer in women with variable survival outcomes. Multiple studies have highlighted GREM-1 as the predominantly overexpressed mRNA in most cancer development with a dominant part

Table-1: Distant metastasis- free survival Hazard ratio (95% CI) and p-value of breast cancer from different studies. All types of breast cancer (ER negative, ER positive, Her-2 positive and triple negative).

	ER positive breast cancer cells	p-value	ER negative breast cancer cells	p-value	All types of breast cancer	p-value
Ren et al. 2019 [30]					1.35 (1.15-1.57)	0.00018
Sung et al. 2020 [29]			2.25 (1.32-3.84)	0.0023		
Park et al. 2020 [17]	1.44 (1-.08)	0.0046	1.99 (1.17-3.37)	0.0092		

in stemness and progression of breast cancer cells. It is directly linked to enhancing multiple pathways necessary for metastasis and cancer cell survival. With all the advancements and clinical trials, the crucial details of GREM-1 remain to be explored and understood better. From the pharmacological aspect, GREM-1 is a possible pharmacodynamics target for many cancers, including breast cancer, due to its multifocal role on a molecular level. However, no drug is designed to target GREM-1 for breast cancer. Further functional research is much needed to fully understand its role in chemotherapy and radiation resistance to increase patients' survival and quality of life.

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Abbreviations

BMP: Bone Morphogenetic Protein

GREM1: Gremlin-1

VEGF: Vascular Endothelial Growth Factor

STAT: Signal Transducer and Activator of Transcription

EMT: Epithelial-Mesenchymal Transition

RFS: Relapse-free survival

DMFS: Distant metastasis-free survival

ERR α: Estrogen-Related Receptor α

TGF- β: Transforming Growth Factor Beta

R-SMAD: Receptor -Regulated Suppressor of mothers against decapentaplegic

ISH: In Situ hybridization

CAF: Cancer-associated fibroblast

ECM: Extra-cellular matrix

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