Pakistan Journal of Life and Social Sciences
www.pjlss.edu.pk

https://doi.org/10.57239/PJLSS-2024-22.1.0052

RESEARCH ARTICLE

Study of Serum Gremlin-1, Galectin-3, and Oxyntomodulin in Women with Breast Cancer

Aya J. Mohammed [1], Entedhar R. Sarhat [2*]

[1] Department of Chemistry, College of Education for Pure Science, Tikrit University, Salah Al-Din, Iraq
[2] Department of Basic Science, College of Dentistry, Tikrit University, Salah Al-Din, Iraq

ARTICLE INFO

Received: Apr 24, 2024

Accepted: June 13, 2024

Keywords

Breast cancer
Gremlin-1
Galectin-3
Oxyntomodulin

***Corresponding Author:**

entedharr@tu.edu.iq

ABSTRACT

The study involved measuring the concentration levels of Gremlin-1, Galectin-3, and oxyntomodulin in the serum of women with breast cancer. It included 60 samples from women with breast cancer and 30 samples from healthy individuals. The results showed that the levels of Gremlin-1 and Galectin-3 are elevated in women with breast cancer, with significant differences with $P \leq 0.0003$ and $P \leq 0.0004$, respectively. Meanwhile, the level of oxyntomodulin is decreased in women with breast cancer compared to the control group, with significant differences ($P \leq 0.007$). Conclusion: The study revealed that the concentration levels of Gremlin-1 and Galectin-3 are significantly elevated in women with breast cancer compared to healthy individuals, while the level of oxyntomodulin is decreased. These findings suggest that these biomarkers may have potential as diagnostic markers for breast cancer. Further research is needed to determine the specific roles of these biomarkers in breast cancer development and progression and to explore their potential as targets for treatment. Overall, this study highlights the importance of biomarker analysis in improving our understanding and management of breast cancer.

INTRODUCTION

Breast cancer (BC) is considered one of the most widespread types of cancer and one of the main causes of death after lung cancer.[1] It represents about 2 million cases each year.[2] Breast cancer develops when its cells grow abnormally, forming tissue masses due to the rapid division and multiplication of cells; these masses are known as tumours. The tumour may be invasive or non-invasive, and it usually begins in the lobules or milk ducts.[3] In the early stages of the disease, patients with breast cancer do not show any symptoms that could lead to diagnosis because the tumour is usually small at the beginning. However, as the tumour grows, it can be felt and detected through its size.[4] Patients with breast cancer suffer from fatigue, anxiety, and depression.[5] Several factors are considered in treating breast cancer, such as the stage of the tumour, the patient's age, and hormones. More than one method can be used to treat breast cancer at the same time. If the tumour is detected early, the breast can be surgically removed, and radiation therapy is used to eliminate any remaining active cancer cells. If the tumour is large and has spread to the lymph nodes, chemotherapy, hormone therapy, and targeted therapy are added to the treatment plan.[6,7]

Gremlin-1 is a novel adipokine, a glycoprotein with a molecular weight of approximately 20.7 KDa that belongs to the Cerberus family in the DNA or cluster of differentiation (CD8).[8,51] It is a member of the cystine knot proteins family, which includes transforming growth factor-β (TGF-β) and vascular endothelial growth factor (VEGF). It also acts as an antagonist for bone morphogenetic proteins (BMPs), especially BMP4 and BMP2, and participates in embryogenesis, bone formation, and organ development.[9] GREM-1 is associated with cellular division and the formation of substructures in lung development and kidney formation. Abnormal expression of GREM-1 in adults is linked to facial and oral clefts, arthritis, spontaneous bone fractures, as well as liver, lung, and kidney fibrosis.[10] Gremlin-1 is a protein that acts as an antagonist to the BMP family. It binds to the BMP2 protein to regulate the BMP signalling pathway, which consists of 184 amino acids and contains a cystine knot, a cysteine-rich area. The first 24 amino acids function as a signal peptide, while the cystine knot is located between amino acids 94-184. The GREM-1 gene is located on chromosome 15q13-q15.[11]

Galectin-3 is found in the lungs, heart, stomach, adrenal gland, ovary, and uterus, and it is the only chimera-type member of the callectin family.[12] Callectin-3 is encoded by the LGALS3 gene located on chromosome 14 at the q21-22 location, which contains 6 exons and 8 introns. The callectin-3 gene contains its regulatory element called galig, which is found in the second intron of the LGALS3 gene.[13] Callectin-3 was formerly known as Carbohydrate-binding protein-35, Mac-2 antigen, IgE-binding protein, and Lectin L-29.[14,50] Callectin-3 is a lectin discovered in the 1980s in cancer cells and has been studied in various organs and diseases associated with it. It consists of 251 amino acid residues and is a glycoprotein that belongs to the lectin protein family, with a molecular weight of 29-35 KDa. It is composed of three domains: the CRD domain, a collagen-like sequence, and the N-terminal domain. The CRD domain consists of 130 amino acids rich in the amino acids Asp-Trp-Gly-Arg, which is a polypeptide binding domain with carbohydrates. The CRD of Galectin-3 interacts with various proteins containing carbohydrates, leading to the activation of different signalling pathways. The collagen-like sequence binds the CRD to the N-terminal domain and consists of nine collagen-like sequences (rich in proline and glycine), which can be cleaved by the metalloproteinase enzymes MMP-2 and MMP-9. The short N-terminal domain is composed of 12 amino acids and contains a serine phosphorylation site to control its transport between the nucleus and cytoplasm.[15]

Oxyntomodulin is a peptide hormone from the incretins family with a molecular weight of approximately 44.499 kDa. It is secreted by enteroendocrine L-cells located at the end of the ileum and colon, and was discovered in 1981.[16] Oxyntomodulin is produced through the conversion of pre-pro-glucagon to pro-glucagon by PCSK1 (Subtilisin/kexin type 1 pro-protein), leading to inhibition of glucagon secretion, reduced caloric intake, increased energy expenditure, and consequently weight loss.[17] Composed of 37 amino acids derived from pro-glucagon, oxyntomodulin is synthesized and secreted by L-cells in the intestines in response to caloric intake.[18] It is named for its ability to regulate gastric acid secretion in the gastric acid glands.[19] Oxyntomodulin binds to and activates both the glucagon receptor (GCGR) and the glucagon-like peptide-1 receptor (GLP-1R), making it a dual agonist for GCGR and GLP-1R receptors.[20] There is no specific receptor for oxyntomodulin; it activates the glucagon receptor, though less effectively than natural glucagon due to the presence of an octapeptide tail, which also allows for activation of the GLP-1 receptor but with less efficacy than natural GLP-1.[21,49] Moreover, the octapeptide tail reduces the clearance of oxyntomodulin from the bloodstream compared to glucagon.[22] Oxyntomodulin is secreted from the intestines along with GLP-1 in response to nutrient intake.[23] It reduces food intake, increases energy expenditure, leads to body weight reduction, and acts as an appetite suppressant.[24]

MATERIALS AND METHODS

After withdrawing 5 mL of blood from women attending the hospital using a 5 mL medical syringe, the blood samples were placed in dry, clean gel tubes and left for 15 minutes at room temperature. Then, the samples were centrifuged for 15 minutes at a speed of 3000 rpm for separation. Serum was drawn using a micropipette, the sediment was discarded, and serum samples were distributed into small amounts in Eppendorf tubes and stored at a temperature of -20°C until testing.

The concentration of GREM-1, Gal-3 and OXM in the serum of women with breast cancer was measured using the ELISA technique with a standard kit produced by Fine Test (Chinese). The method included the steps provided by the manufacturer.

The results of the study were statistically analyzed using the SPSS statistical program, utilizing the ANOVA test. The mean values of the parameters were compared using Duncan's multiple range test at a significance level of ($P < 0.05$) significant, ($P > 0.05$) not significant. Results were expressed in percentage ratios and the mean ± standard deviation. Microsoft Excel was also used to create graphs, charts, and tables.

RESULTS

The study included 60 cases of women with breast cancer. Additionally, it included 30 samples from a control group, used as a comparative group, with ages ranging from 20 to 65 years.

The results showed that the gremlin concentration in women with breast cancer was 5.45 ± 1.93 ng/ml, and the concentration in the control group was 4.22 ± 0.649 ng/ml. Statistical analysis revealed significant differences at the probability level of $P \leq 0.0003$, indicating an increased concentration level of gremlin-1 in women with breast cancer compared to the control group (Table 1).

The results indicated that the galectin-3 concentration in women with breast cancer was 415.1 ± 61.4 pg/ml, and the concentration in the control group was 315.5 ± 49.2 pg/ml. Upon conducting statistical analysis, significant differences were observed at the probability level of $P \leq 0.0004$, indicating an increased concentration of galectin-3 in women with breast cancer compared to the control group (Table 1).

The results showed that the oxyntomodulin concentration in women with breast cancer was 149.0 ± 27.7 ng/ml, and the concentration in the control group was 177.6 ± 31.0 ng/ml. Statistical analysis indicated significant differences at the probability level of $P \leq 0.007$, with a decrease in the oxyntomodulin level in women with breast cancer compared to the control group (Table 1).

Table 1. Serum GREM-1, Gal-3, and OXM concentrations of women with breast cancer and healthy women

Parameters	Control (*n*=30)	Patients (*n*=60)	*P* value
GREM-1 (ng/ml)	4.22 ± 0.649	.45±1.93	0.0003
Gal-3 (pg/ml)	315.5 ± 49.2	415.1 ± 61.4	0.0004
OXM (ng/ml)	177.6 ± 31	149.0 ± 27.7	0.007

Data expressed as mean ± standard deviation, Unpaired two sample t-test used with significance considered at a p-value of 0.05

The results we obtained were analyzed by performing the Receiver Operating Characteristic (ROC) curve analysis, which is based on several values including:

ROC curve for gremlin-1 level: The AUC value is 0.869 and the Cut-off value is greater than 4.2185 ng/ml, with a sensitivity of 85.0%, while the specificity was 76.7%. The study results suggest that the accuracy of the AUC was excellent for the level of Gremlin-1 and that a Cut-off value higher than 4.2185 ng/ml could be considered a diagnostic indicator of the disease with very good sensitivity.

ROC curve for galectin-3 level: The AUC is 0.787 and the Cut-off value is higher than 385.4677 pg/ml, with a sensitivity of 51.7%, while the specificity was 93.3%. The current results show that the accuracy of the AUC was excellent and that the Galectin-3 value may serve as an important diagnostic indicator when its concentration exceeds 385.4677 pg/ml, with acceptable sensitivity and excellent specificity.

3.4.3. ROC curve for oxyntomodulin level: The AUC is 0.653 and the Cut-off value is less than or equal to 187.9832 ng/ml, with a sensitivity of 98.3%, while the specificity was 46.7%. The study results suggest that the accuracy of the AUC was excellent for the level of oxyntomodulin and that a value lower than 187.9832 ng/ml could be considered an important diagnostic indicator of breast cancer with excellent sensitivity (Figure 1).

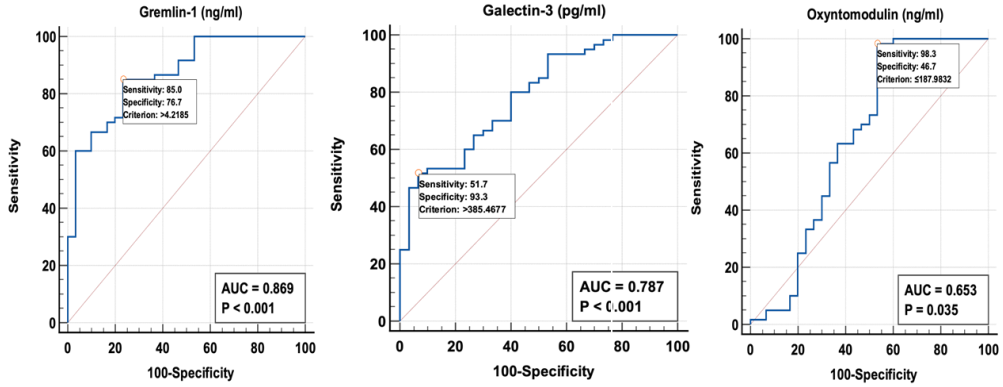

Figure 1. ROC analysis of measured parameters.

DISCUSSION

The elevated levels of Gremlin-1 and Galectin-3 in women with breast cancer suggest their potential as diagnostic markers, with their significant differences demonstrating their potential for early detection. On the other hand, the decreased levels of oxyntomodulin in women with breast cancer may indicate its potential as a prognostic marker for disease progression.

The results indicated that the concentration level of Gremlin-1 increased in women with breast cancer compared to the control group. These results are in agreement with the findings of Park et al. (2020), and his group, which demonstrated that the level of Gremlin-1 is elevated in cells and tissues of women with breast cancer, particularly in those with estrogen receptor-negative breast tumors.[25,46] Decreased expression of Gremlin-1 protein inhibits the proliferation of breast cancer cells and tumour growth, while increased expression of the protein enhances the growth potential, proliferation, and metastatic ability of cancer cells. These findings are also consistent with the results of Kim et al. (2020) and his team, which studied the role of Gremlin-1 in glycolysis in breast cancer

cells and the processes it mediates.[26,52] Ghanbari et al. (2019) study showed that breast cancer cells with high Gremlin-1 secretion increase glucose uptake and lactate production through glycolysis, leading to a decrease in pH within the tumour environment, a distinctive hallmark of cancer growth and development, such as the oxidative tumour environment, tumour growth, and dissemination.[27] To date, studies explaining the effect of Gremlin-1 on the growth and spread of cancer cells are limited. Some studies have found that transforming growth factor-β (TGF-β) increases the level of Gremlin-1. Additionally, activation of nuclear factor-kB signalling by reactive oxygen species also increases the level of Gremlin-1.[28] Gremlin-1 is also widely expressed in cells and tissues associated with tumours and is one of the genes that is markedly elevated in the connective tissue associated with breast tumours, especially in cancer-associated fibroblasts (CAFs) in basal cell carcinoma. Some studies have found that Gremlin-1 derived from CAFs promotes the spread of breast cancer cells.[29]

The study's findings are in line with the results of Ibrahim (2023)[30] and his group, Shafiq et al. (2020)[31] and Patel et al. (2021),[32] which showed that the level of galectin-3 is elevated in women with breast cancer. Galectin-3 is a carbohydrate-binding protein soluble in water and influences various cellular processes.[33] The levels of galectin-3 depend on the cell type and stimulants, and an increase in galectin-3 protein concentration in breast cancer cells makes them resistant to chemotherapy.[34,47] Conditions of stress such as hypoxia, poor nutrition, and the accumulation of galectin-3 in the cytoplasm lead to increased concentrations of galectin-3 in breast cancer. Galectin-3 is produced in the nucleus, cytoplasm, and extracellular matrix and is of significant importance in breast cancer because it enhances the interaction between cellular components and the extracellular matrix, which increases tumour spread.[35,45] Galectin-3 has multiple biological functions; it can be distributed inside and outside the cells and includes abnormal cell division, migration, angiogenesis, cell interaction, differentiation, tumour progression, apoptosis, adhesion, and epithelial-mesenchymal transition.[36,43]

The results demonstrated that the concentration level of oxyntomodulin decreases in women with breast cancer compared to the control group. These findings align with the results of Faiq (2020)[37,44] which indicated a decrease in oxyntomodulin levels in women with breast cancer in comparison to the control group.

Oxyntomodulin is produced from pre-proglucagon and is a peptide that can bind with the glucagon receptor (GCGR) and the glucagon-like peptide-1 receptor (GLP-1R).[38] Oxyntomodulin consists of the original oxyntomodulin, derived forms, and oxyntomodulin analogues, containing the amino acid sequence H-S-Q-G-T-F-T-S-D-Y-S-K-Y-L-D-S-R-R-A-Q-D-F-V-Q-W-L-M-N-T-K-R-N-R-N-N-I-A.[39,48] Oxyntomodulin has been studied for the treatment of obesity, hyperlipidemia, and fatty liver disease. Researchers found that derived oxyntomodulin is more effective than the original form in reducing blood lipid levels; therefore, it can be used as an effective treatment for hyperlipidemia. Derived oxyntomodulin also has a high capacity to activate GLP-1R, assisting in lowering blood cholesterol levels and triglycerides that increase due to a high-fat diet.[40] Oxyntomodulin is used in the treatment of diabetes, where oxyntomodulin analogues show a greater ability to activate the GLP-1R, promoting the expansion of beta cells, increasing insulin secretion, and thus reducing blood sugar levels. Oxyntomodulin analogues also help reduce body weight and suppress appetite, which in turn helps maintain normal blood glucose levels. Oxyntomodulin is used for weight reduction by decreasing food intake and aiding in fat breakdown.[41] Hence, oxyntomodulin plays an important role in reducing the risk of breast cancer since obesity is one of the risk factors for the disease, in the future, oxyntomodulin may be used as a treatment in controlling breast cancer as it regulates body weight and is one of the important factors in preventing obesity.[42]

CONCLUSION

In conclusion, the serum concentration levels of Gremlin-1, Galectin-3, and Oxyntomodulin show significant potential as biomarkers for breast cancer. Higher levels of Gremlin-1 and Galectin-3 alongside lower levels of Oxyntomodulin in patients compared to a healthy control group provide a foundation for using these biomarkers in diagnosis. The Receiver Operating Characteristic (ROC) curve analysis confirms the diagnostic accuracy of these biomarkers, with Gremlin-1 and Galectin-3 demonstrating high AUC values and Oxyntomodulin showing very high sensitivity. These biomarkers, therefore, could become essential tools in early detection and could also serve as indicators of disease progression. The study also suggests therapeutic implications, particularly with the derived forms of Oxyntomodulin, which have been effective in metabolic regulation—a significant factor considering the link between obesity and breast cancer. This points to the potential for targeted therapeutic interventions that take advantage of these findings. Moving forward, further exploration into the roles of these biomarkers could enhance our understanding of breast cancer's prognosis and treatment, ultimately leading to improved patient care.

References

1. Faruqui N, Yousuf MA, Whaiduzzaman M, Azad AK, Barros A, Moni MA. LungNet: A hybrid deep-CNN model for lung cancer diagnosis using CT and wearable sensor-based medical IoT data. Computers in Biology and Medicine. 2021 Dec 1;139:104961.
2. Sarhat ER, Abbas MQ, Ali NH, Wadi SA, Sarhat TR. Evaluation of ceruloplasmin, sialic acid and liver function for women with breast cancer. InAIP Conference Proceedings 2022 Nov 8 (Vol. 2394, No. 1). AIP Publishing.
3. Rubagumya F, Makori K, Dharsee N, Tausi M. Thyroid function post supraclavicular lymph node irradiation in patients with breast cancer. RMJ. 2019 Mar 1;80:1.
4. Aziz Z, Sarhat E, Zaidan Z. Estimation of serum ferroportin and liver enzymes in breast cancer patients. Georgian medical news. 2023 Jun(339):37-41.
5. Al-Barwani R, Sarhat E. Breast cancer-modulated omentin and vaspin plasma levels. Georgian Medical News. 2024 Feb 1(347):66-9.
6. Abd-Alqader F, Sarhat E, Zaidan Z. Evaluation of the role of coenzyme Q 10 in the blood of breast cancer women. Georgian Medical News. 2023 May 1(338):91-5.
7. Merkhan MM, Faisal IM, Alsaleem DZ, Shindala OM, Almukhtar HM, Thanoon IA. Immunodepressant and oxidant potential of standard leukaemia drug regimen. International Journal of Research in Pharmaceutical Sciences. 2020;11(4):1-4.
8. Allow S, Sarhat E. Metformin effects on blood levels of gremlin-1 in polycystic ovarian women. Georgian medical news. 2023 Apr 1(337):51-5.
9. Li R, Zhou H, Li M, Mai Q, Fu Z, Jiang Y, Li C, Gao Y, Fan Y, Wu K, Costa CD. Gremlin-1 promotes colorectal cancer cell metastasis by activating ATF6 and inhibiting ATF4 pathways. Cells. 2022 Jul 7;11(14):2136.
10. Mahmood DA, Sarhat ER, Sulaiman YA, Abass KS. Evaluation of liver function tests in patients with psoriasis. Revista Latinoamericana de Hipertension. 2022;17(6).
11. Aoshima Y, Enomoto Y, Muto S, Meguro S, Kawasaki H, Kosugi I, Fujisawa T, Enomoto N, Inui N, Nakamura Y, Suda T. Gremlin-1 for the differential diagnosis of idiopathic pulmonary fibrosis versus other interstitial lung diseases: a clinical and pathophysiological analysis. Lung. 2021 Jun;199:289-98.

12. Nio-Kobayashi J. Tissue-and cell-specific localization of galectins, β-galactose-binding animal lectins, and their potential functions in health and disease. Anatomical Science International. 2017 Jan;92:25-36.
13. Mahmood DA, Sarhat ER, Sulaiman YA, Abass KS. Relationship between Paraoxonase and Malondialdehyde as a marker of oxidative stress in patients with psoriasis. Revista Latinoamericana de Hipertension. 2022;17(6).
14. Sciacchitano S, Lavra L, Morgante A, Ulivieri A, Magi F, De Francesco GP, Bellotti C, Salehi LB, Ricci A. Galectin-3: one molecule for an alphabet of diseases, from A to Z. International journal of molecular sciences. 2018 Jan 26;19(2):379.
15. Boutin L, Dépret F, Gayat E, Legrand M, Chadjichristos CE. Galectin-3 in kidney diseases: from an old protein to a new therapeutic target. International journal of molecular sciences. 2022 Mar 14;23(6):3124.
16. Masadah R, Ekawardhani S, Putra RE, Ikram D, Faruk M. Ethanolic Extract of Propolis from Tetragonula laeviceps: Selective Cytotoxicity for MCF-7 Breast Cancer cells. Pharmacognosy Journal. 2023;15(6):1177-1180.
17. Scott R, Minnion J, Tan T, Bloom S. Oxyntomodulin analogue increases energy expenditure via the glucagon receptor. Peptides. 2018 Jun 1;104:70-7.
18. Naser SI, Fakhree NK, Mhaibes SH. Regulation of Appetite and Satiety by Gastrointestinal Peptides. Iraqi Journal of Pharmaceutical Sciences. 2021;30(1).
19. Pocai A. Action and therapeutic potential of oxyntomodulin. Molecular metabolism. 2014 Jun 1;3(3):241-51.
20. Wang J, Liu C, Yang H, Ma T, Liu Y, Chen F. Intramolecularly lactam stapled oxyntomodulin analogues inhibit cancer cell proliferation in vitro. Chinese Chemical Letters. 2023 Apr 1;34(4):107920.
21. Jonnakuti VS, Stanescu DE, De Leon DD. Developmental Origins and Roles of Intestinal Enteroendocrine Hormones. InMaternal-Fetal and Neonatal Endocrinology 2020 Jan 1 (pp. 599-610). Academic Press.
22. Azubuike SO, Beeler D. Novel breast cancer risk factors in Nigeria: the findings of Nigerian breast cancer risk factor study. Rawanda Medical Journal. 2023;80(3):24-33.
23. Bharmal SH, Cho J, Stuart CE, Ramos GC, Ko J, Petrov MS. Oxyntomodulin may distinguish new-onset diabetes after acute pancreatitis from type 2 diabetes. Clinical and translational gastroenterology. 2020 Feb 1;11(2):e00132.
24. Dakin CL, Gunn I, Small CJ, Edwards CM, Hay DL, Smith DM, Ghatei MA, Bloom SR. Oxyntomodulin inhibits food intake in the rat. Endocrinology. 2001 Oct 1;142(10):4244-50.
25. Park SA, Sung NJ, Choi BJ, Kim W, Kim SH, Surh YJ. Gremlin-1 augments the oestrogen-related receptor α signalling through EGFR activation: implications for the progression of breast cancer. British journal of cancer. 2020 Sep 15;123(6):988-99.
26. Kim NH, Sung NJ, Youn HS, Park SA. Gremlin-1 activates Akt/STAT3 signaling, which increases the glycolysis rate in breast cancer cells. Biochemical and Biophysical Research Communications. 2020 Dec 17;533(4):1378-84.
27. Movahed ZG, Rastegari-Pouyani M, Hossein Mohammadi M, Mansouri K. Cancer cells change their glucose metabolism to overcome increased ROS: One step from cancer cell to cancer stem cell?. Biomedicine & Pharmacotherapy. 2019 Apr 1;112:108690.
28. Chang SH, Mori D, Kobayashi H, Mori Y, Nakamoto H, Okada K, Taniguchi Y, Sugita S, Yano F, Chung UI, Kim-Kaneyama JR. Excessive mechanical loading promotes osteoarthritis through the gremlin-1–NF-κB pathway. Nature communications. 2019 Mar 29;10(1):1442.
29. Ren J, Smid M, Iaria J, Salvatori DC, van Dam H, Zhu HJ, Martens JW, Ten Dijke P. Cancer-associated fibroblast-derived Gremlin 1 promotes breast cancer progression. Breast cancer research. 2019 Dec;21:1-9.

30. Ibrahim AA, Ahmed SN. Study of Serum Adiponectin, Leptin, and Galectin-3 in Breast Cancer. Diyala Journal of Medicine. 2023 Apr 5;24(1):185-96.
31. Shafiq A, Moore J, Suleman A, Faiz S, Farooq O, Arshad A, Tehseen M, Zafar A, Ali SH, Din NU, Loya A. Elevated soluble galectin-3 as a marker of chemotherapy efficacy in breast cancer patients: A prospective study. International journal of breast cancer. 2020 Mar 14;2020.
32. Patel SR, Herrmann J, Vierkant RA, Olson JE, Couch FJ, Hazim A, Sloan JA, Loprinzi CL, Ruddy KJ. N-terminal pro brain natriuretic peptide, sST2, and galectin-3 levels in breast cancer survivors. Journal of Clinical Medicine. 2021 Jul 27;10(15):3313.
33. Hsu DK, Liu FT. Regulation of cellular homeostasis by galectins. Glycoconjugate journal. 2002 Jan;19:507-15.
34. Takenaka Y, Fukumori T, Yoshii T, Oka N, Inohara H, Kim HR, Bresalier RS, Raz A. Nuclear export of phosphorylated galectin-3 regulates its antiapoptotic activity in response to chemotherapeutic drugs. Molecular and cellular biology. 2004 May 1;24(10):4395-406.
35. Pereira JX, Dos Santos SN, Pereira TC, Cabanel M, Chammas R, de Oliveira FL, Bernardes ES, El-Cheikh MC. Galectin-3 regulates the expression of tumor glycosaminoglycans and increases the metastatic potential of breast cancer. Journal of oncology. 2019 Dec 17;2019.
36. Aljurany AN, Al Samarrai OR. Evaluation of galectin-3 and ferritin with liver enzymes in women with breast cancer. International Journal of Health Sciences.(III):6404-14.
37. Faiq. Maryam Abdul Ameer (2020). Evaluation serum levels of vimentin and oxyntomodulin with som biochemical parameters in women with breast cancer. Tikrit University. College of Medicine.
38. Cai X, Li C, Zhou J, Dai Y, Avraham Y, Sun L, Liu C, Tong J, Wang Y, Bi X, He L. Novel glucagon-and OXM-based peptides acting through glucagon and GLP-1 receptors with body weight reduction and anti-diabetic properties. Bioorganic Chemistry. 2020 Jan 1;95:103538.
39. Sarhat ER, Wadi SA, Mahmood AR. Effect of ethanolic extraction of moringa oleifera on paraoxonase and arylesterase enzyme activity in high fat diet-induced obesity in rats. Research Journal of Pharmacy and Technology. 2018;11(10):4601-4.
40. Yang PY, Zou H, Amso Z, Lee C, Huang D, Woods AK, Nguyen-Tran VT, Schultz PG, Shen W. New generation oxyntomodulin peptides with improved pharmacokinetic profiles exhibit weight reducing and anti-steatotic properties in mice. Bioconjugate Chemistry. 2020 Apr 3;31(4):1167-76.
41. Kim JS, Kim DJ, Lee SH, youb Jung S, Kwon SC, inventors; Hanmi Pharmaceutical Co Ltd, assignee. Composition for treating diabetes or diabesity comprising oxyntomodulin analog. United States patent US 10,550,168. 2020 Feb 4.
42. Mahmmod M, Sarhat E. Hepcidin and Ferritin Modulated in Obese Male. Georgian Medical News. 2023 Nov 1(344):114-8.
43. Jam, F. A., Rauf, A. S., Husnain, I., Bilal, H. Z., Yasir, A., & Mashood, M. (2014). Identify factors affecting the management of political behavior among bank staff. African Journal of Business Management, 5(23), 9896-9904.
44. Jam, F. A., Singh, S. K. G., Ng, B., & Aziz, N. (2018). The interactive effect of uncertainty avoidance cultural values and leadership styles on open service innovation: A look at malaysian healthcare sector. International Journal of Business and Administrative Studies, 4(5), 208-223.
45. Alneyadi, S., Abulibdeh, E., & Wardat, Y. (2023b). The impact of digital environment vs. traditional method on literacy skills; reading and writing of Emirati fourth graders. Sustainability, 15(4), 3418. https://doi.org/10.3390/su15043418
46. Alneyadi, S., Wardat, Y., Alshannag, Q., & Abu-Al-Aish, A. (2023a). The effect of using smart e-learning app on the academic achievement of eighth-grade students. Eurasia Journal of Mathematics, Science and Technology Education, 19(4), em2248. https://doi.org/10.29333/ejmste/13067

47. Wardat, Y., Tashtoush, M., AlAli, R., & Saleh, S. (2024). Artificial intelligence in education: Mathematics teachers' perspectives, practices and challenges. Iraqi Journal for Computer Science and Mathematics, 5(1), 60-77.
48. Zakariya, Y. F., & Wardat, Y. (2023). Job satisfaction of mathematics teachers: An empirical investigation to quantify the contributions of teacher self-efficacy and teacher motivation to teach. Mathematics Education Research Journal. https://doi.org/10.1007/s13394-023-00475-9
49. Kanval, N., Ihsan, H., Irum, S., & Ambreen, I. (2024). Human Capital Formation, Foreign Direct Investment Inflows, and Economic Growth: A Way Forward to Achieve Sustainable Development. Journal of Management Practices, Humanities and Social Sciences, 8(3), 48-61
50. Jarrah, A. M., Wardat, Y., & Gningue, S. (2022b). Misconception on addition and subtraction of fractions in seventh-grade middle school students. Eurasia Journal of Mathematics, Science and Technology Education, 18(6), em2115. https://doi.org/10.29333/ejmste/12070
51. Tashtoush, M. A., AlAli, R., Wardat, Y., Alshraifin, N., & Toubat, H. (2023b). The impact of information and communication technologies (ICT)-based education on the mathematics academic enthusiasm. Journal of Educational and Social Research, 13(3), 284. https://doi.org/10.36941/jesr-2023-0077
52. Tashtoush, M. A., Wardat, Y., & Elsayed, A. M. (2023a). Mathematics distance learning and learning loss during COVID-19 pandemic: Teachers' perspectives. Journal of Higher Education Theory and Practice, 23(5). https://doi.org/10.33423/jhetp.v23i5.5933