Caspian Journal of Internal Medicine 2024 (Autumn); 15(4): 706-712
DOI: 10.22088/cjim.15.4.706

Short communication

Insulin-like growth factor (IGF) levels in pre-treatment plasma identifying breast cancer: A case control study

Sarvin Alizadeh Sadighi (MD) [1]
Nematollah Rostami (MD) [2*]
Maryam Tohidi (MD) [3]
Mahbobeh Mashayekhi (MD) [4]

1. Department of Internal Medicine, Clinical Research Development Center at Modarres Hospital, Shahid Beheshti University of Medical Science, Tehran, Iran
2. Department of Hematology, Modarres Hospital, Shahid Beheshti University of Medical Science, Tehran, Iran
3. Prevention of Metabolic Disorders Research Center, Research Institute for Endocrine Sciences, Shahid Beheshti University of Medical Sciences, Tehran, Iran
4. Department of Endocrinology, Modarres Hospital, Shahid Beheshti University of Medical Sciences, Tehran, Iran

*** Correspondence:**
Nematollah Rostami, Department of Internal Medicine, Clinical Research Development Center at Modarres Hospital, Shahid Beheshti University of Medical Sciences, Tehran, Iran

E-mail: dnrostami@gmail.com
Tel: + 98 4133340905

Received: 17 May 2023
Revised: 3 Dec 2023
Accepted: 5 Dec 2023
Published: 7 Sep 2024

Abstract

Background: Diabetes (primarily type 2) is linked to a higher risk of breast cancer. Insulin-like growth factor (IGF) is one of the most important factors that affects mitosis and thus inhibits apoptosis. The purpose of this study was to compare the pre-treatment insulin-like growth factor (IGF) levels in breast cancer against normal population.

Methods: In this case-control study, 60 patients with breast cancer and 60 healthy controls were enrolled in 2017 and 2018 at Tehran's Shahid-Modarres Hospital. In this study, the blood sugar of the patients was examined before entering the study, and the age of the patients was also within the age limit of 18 to 70 years. They were studied to determine the relationship between insulin-like growth factor (ELISA method) and breast cancer.

Results: Both groups have similar IGF-1 levels (Ctrl and Case) (P= 0.188). But, IGF-2 levels were significantly higher in breast cancer patients (373.4 vs. 317.3 ng/ml), (P=0.0001).

Conclusion: According to our study, IGF-2 may serve as a prognostic biomarker and potential therapeutic target for breast cancer. However, further investigation is needed to validate this claim.

Keywords: Breast cancer, IGF-1, IGF-2, Diabetes, ELISA.

Citation:
Alizadeh Sadighi S, Rostami N, Tohidi M, Mashayekhi M**.** Insulin-like growth factor (IGF) levels in pre-treatment plasma identifying breast cancer: A case control study. Caspian J Intern Med 2024; 15(4): 706-712.

Breast cancer is a common malignancy worldwide with increasing trend despite medical adventures (1, 2). It is the most prevalent type of cancer in women, affecting almost one in every eight female patients in the United States (3-5). Hormones including estrogen and progesterone and insulin-like growth factor (IGF-1) are contributing factors for breast cancer due to proliferative effects (5, 6). IGF are effective in growth, proliferation, and differentiation by cellular effects (1, 4, 7).

This system has two superficial cellular receptors (IGF-1R, IGF-2R), two legends (IGF1, IGF-2) and six binding proteins (IGFBP) and there is membranous receptor for metabolic effects especially with mitogenic and anti-apoptotic properties (1, 4, 7, 8). Certain types of cancer, such as colon, prostate, melanoma, lung, breast, osteosarcoma, and pediatric malignancies, have been linked to elevated IGF-1 levels (4, 7, 8). Additional associations are demonstrated between elevated IGF-2 and colorectal cancer, as well as between IGFBP-3 inhibition and osteosarcoma (8, 9).

Also IGF pathway is shown to be related to increased morbidity and mortality of cancers due to chemotherapy and radiotherapy resistance and high morbidity and reduced survival (4, 10). Increased IGF-1R is shown to be related to tamoxifen resistance in breast cancer cases (11). It may be related to decreased efficacy of trastuzumab in breast cancer cases with some degrees (4). It is not yet clear that increased expression of IGF-1 gene is directly related to breast cancer risk or it is due to menopausal status, estrogen receptor, and IGFBP-3 (8). Also, definite prognostic role of IGF-1 in breast cancer cases is not yet understood (10-12).

Although the IGFBP-3 is decreased in breast cancer cases, it may be seen as gene over-expression in highly invasive breast malignancies (12). The purpose of this study was to gain a better understanding of how insulin-like growth factor (IGF) levels in pre-treatment plasma serve as a biomarker for breast cancer.

Methods

Patients, blood sample: In this case-control study, 60 randomly-selected cases with breast cancer and 60 control subjects attending to Shahid-Modarres Hospital in Tehran, Iran in February 2017 and Jeanery 2018 were enrolled.

Patients recruited to this longitudinal study provided informed written consent. Inclusion criteria were age range from 18 to 70 years, established breast cancer cases by imaging and pathology in case group. Among these women (n=60), n=26 premenopausal and n=34 postmenopausal) were found. The exclusion criteria were non-established breast cancer in case group and undetermined serum level of IGF-1 and IGF-2. The study was approved by local Ethics Committee in Shahid-Beheshti University of Medical Sciences. Two groups were matched for age, body mass index, and diabetes mellitus history. The blood sample was obtained by venous sampling and it was rapidly centrifuged. The extracted plasma was reserved in -80 centigrade degree till all samples were collected.

Measures: Patients with diabetes were excluded from the study. BMI was determined through the measurement of height and weight at the baseline assessment. Participants in the study groups did not receive any treatment, including surgery, chemotherapy, or radiation therapy, until sampling.

ELISA investigations: Serum IGF-1 and IGF-2 levels were measured by double antibody sandwich ELISA according to the manufacturer's instructions (DY291, DY871, R&D Systems, USA).

Briefly, 96-well microtiter plates were coated with 100 µL/well of capture antibody (mouse anti-human IGF-1 or IGF-2, 4.0 µg/mL) overnight at 4°C. After blocking with 3% BSA, 100 µl of the test tests 1 100 weakened in 1 BSA was included and brooded for 2 h at room temperature. Along these lines, 100 l well of the discovery counter acting agent biotinylated goat anti human IGF 1 150 ng ml or IGF 2 400 ng ml was included and brooded for 2 h at room temperature. Following, 100 l well of Streptavidin HRP 1 200 was included and brooded for 20 min at room temperature. At long last, the substrate tetramethylbenzidine arrangement was included, and the response was ended utilizing 2 N H_2SO_4 and examined at an OD of 450 nm. Each test included a standard control CV 12.

Statistical Data Analyzes: All data analyses were performed using GraphPad Prism v 8. T-tests were used to assess differences in IGF-1 and IGF-2 serum levels between study patients and healthy controls. A p-value is indicated as statistically significant at the level of < 0.05.

Results

Patient characteristics: The clinical and molecular characteristics of breast cancer patients are summarized in tables 1 and 2. The mean age was 51.35 ± 10.88 and 47.76 ± 5.71 in the case and control groups, respectively ($p > 0.05$). The mean BMI was 28.28 ± 4.95 and 27.38 ± 4.01 in the case and control groups, respectively ($p> 0.05$).

Serum IGF1 and IGF2 levels: As shown in figures 1 and 2, both groups have similar IGF-1 levels (Ctrl and Case) (P= 0.188), and the IGF-2 was higher in case group and the difference was statistically significant (P=0.001). There was no significant difference according to age for IGF-1 but about the IGF-2 levels, in ages older than 60 years there was significantly higher levels (P=0.037).

Table1. Clinico-pathological characteristics of patients

Parameter	category	Number
Stage	I	31
	II	12
	III	17
Grade	I	35
	II	20
	III	3
	Unknown	2

Parameter	category	Number
N staging	N0	30
	Ni	19
	N2	7
	N3	3
	Nx	1
T Staging	T1	35
	T2	19
	T3	4
	T4	2

Table2. Molecular characteristics of patients

Molecular subtypes	Stage	TNM	Pathology	HER-2	PR	ER	case
							1
Luminal A	IV	T1N2M1		-	+	+	**2**
TN				-	-	-	**3**
TN	llB	T2N1M0		-	-	-	**4**
Luminal A	llA	T1N1M0	Invasive ductal	-	+	+	**5**
TN	llA	T2N0M0	Invasive ductal	-	-	-	**6**
Luminal B	llA	T2N0M0		+	+	+	**7**
Luminal A				-	+	+	**8**
Luminal B			Invasive ductal	+	+	+	**9**
Luminal A	IV	T2N1M1		-	+	+	**10**
Luminal B	lllA	T1N2M0		+	+	+	**11**
Luminal A	lA	T1N0M0	Invasive lobular	-	+	+	**12**
Luminal A	llB	T2N1M0	Invasive ductal	-	+	+	**13**
							14
							15
Luminal A	llB	T2N1M0	Invasive ductal	-	+	+	**16**
HER2	lllA	T2N2M0	Invasive adenocarcinoma	+	-	-	**17**
Luminal A			Invasive lobular	-	+	+	**18**
Luminal A	llA	T2N0M0	Invasive ductal	-	+	+	**19**
Luminal A	llB	T2N1M0		-	+	+	**20**
HER2	llA	T2N0M0	Invasive ductal	+	-	-	**21**
Luminal A	lV	T2N0M1		-	+	+	**22**
Luminal A	IV	T1N1M1		-	+	+	**23**
HER2	IV	T1N2M1		+	-	-	**24**
Luminal B	IV	T2N3M1	Invasive ductal	+	-	+	**25**

Molecular subtypes	Stage	TNM	Pathology	HER-2	PR	ER	case
	IV	T3N1M1	Papillary cancer				**26**
Luminal B	ll B	T2N1M0		+	+	+	**27**
Luminal A	llB	T2N1M0	Invasive ductal	-	+	+	**28**
	llA	T2N0M0		-	-	+	**29**
							30
Luminal A	llB	T2N1M0		-	+	+	**31**
Luminal A	llA	T1N1M0		-	+	+	**32**
Luminal A	llB	T2N1M0		-	+	+	**33**
Luminal A	IV	T2N2M1		-	+	+	**34**
Luminal A			Intraductal carcinoma	-	+	+	**35**
							36
Luminal A				-	+	+	**37**
							38
	llA	T2N0M0					**39**
TN	llA	T2N0M0		-	-	-	**40**
							41
Luminal A	llA	T1N1M0	Invasive ductal	-	+	+	**42**
Luminal A			Invasive ductal	-	+	+	**43**
Luminal A	lllA	T1N2M0	Invasive ductal	-	+	+	**44**
Luminal A	llB	T2N1M0		-	+	+	**45**
Luminal A	IV	T3N2M1		-	+	+	**46**
Luminal B	lV	T2N1M2		+	+	+	**47**
HER2			Invasive ductal	+	-	-	**48**
Luminal A	llB	T2N1M0	Invasive ductal	-	+	+	**49**
Luminal A	llB	T2N1M0	Invasive ductal	-	-	+	**50**
TN	llB	T2N1M0		-	-	-	**51**
TN	IV	T2N3M2	Invasive ductal	-	-	-	**52**
Luminal B	IV	T3N2M1		+	+	+	**53**
Luminal B	lllA	T2N2M0		+	+	+	**54**
Luminal B	llB	T2N1M0		+	+	+	**55**
Luminal A	llA	T2N0M0		-	+	+	**56**
Luminal B	IV	T1N1M1	Invasive ductal	+	+	+	**57**
Luminal B			Invasive ductal	+	+	+	**58**
Luminal A			Invasive ductal	-	+	+	**59**
HER2	llB	T2N1M0	Invasive ductal	+	-	-	**60**

Caspian Journal of Internal Medicine 2024 (Autumn); 15(4): 706-712
Alizadeh Sadighi S, et al.

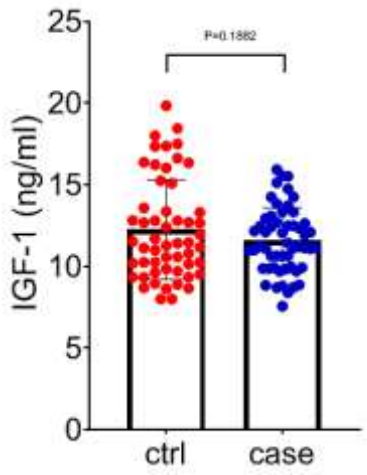

Figure 1. Serum level of IGF-1 across the groups

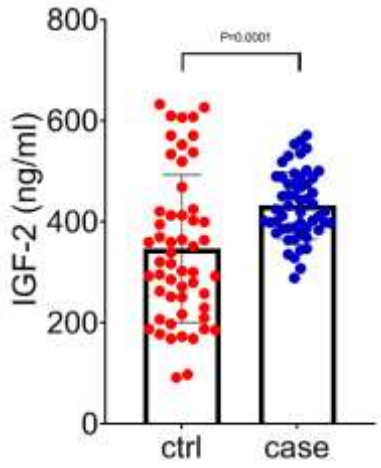

Figure 2. Serum level of IGF-2 across the groups

Discussion

Peptide hormones in IGF class can regulate the growth by cellular stimulation and inhibition of apoptosis that can affect the tumor growth pattern (13, 14). Since breast cancer has shown an increasing trend in recent years, in this study the serum levels of IGF-1 and IGF-2 were compared between breast cancer cases and normal control subjects. This study showed a significant increase in IGF-2 concentration in serum levels of patients with breast cancer compared to healthy age-matched women. Also, IGF-1 level was higher in case group but without significant difference. IGF-1 is known to promote cancer development by inhibiting apoptosis and stimulating cell proliferation. In the current study, have reported a positive association between circulating IGF-1 levels and various primary cancers, including breast cancer, colon cancer, and prostate cancer (13-15).

The loss of imprinting of IGF2 and an overexpression of this growth factor gene have been reported in a wide range of cancer (16, 17). Ulrick Espelund et al (5) assessed 43 early-stage breast cancer cases and reported higher serum IGF-1 and IGF-2 levels versus healthy subjects. In this study, IGF was reported as a contributing factor for progression of breast cancer. As a limitation in our study, the stage of malignancy was not considered. But it may be also assessed by further studies. The relationship between IGF-1 and IGFBP-3 and breast cancer risk was examined by the Endogenous Hormones and Breast Cancer Collaboration Group (6).

The authors reported that IGF-1 level was related to age in first pregnancy and height. Also, it has reverse correlation with menarche age and the interval with menopause and its level is higher in overweight cases and alcohol users. They also found a significant difference between IGF-1 levels across the groups but IGFBP-3 was similar (18). Studies have shown that IGF-1 gene expression increases in breast cancer. IGF-1 and IGF-2 effects in growth, proliferation and tumor genesis is mediated by IGF-1 receptor (19-21). IGFBP3 inhibit IGF-1 effects by preventing its binding to a surface receptor (IGF-1R) and interferes with IGF-1 mitoses action (22-24). Figure 3 briefly illustrates the role of IGF-1 and IGF-2 in tumor formation. IGF-1 and IGF-2 both stimulate the IGF-1 receptor and activate its downstream signal. IGF-1 receptor affects metabolism, proliferation, cell growth and apoptosis in two different ways. IGF1 receptor is composed of two αβ dimers which associate to form heterotetrameric complexes (25-28).

The αβ dimers are connected by disulfide bonds, and two further dimers are likewise connected via disulfide bonds to create the tetramer. The cytoplasmic component of the subunit interacts with IRS proteins, which are important intracellular mediators of insulin/IGF signaling, whereas the β subunit spans the membrane. The α subunit is the receptor's extracellular portion. (29, 30). IGF1 receptor signaling is mediated by a complex, highly integrated network that controls several processes. Two pathways are activated by IGF1 receptor signaling, the ATK/PI3K pathway and the ERK/MAPK pathway, which are important in several cellular processes such as metabolism, cell growth, proliferation, and apoptosis. The β subunits are phosphorylated when IGF1/IGF2 binds to the IGF1 receptor, and the receptor tyrosine kinase then phosphorylates IRS proteins on their tyrosine residues. (31, 32). Overall, this study showed significant concentrations IGF-1 and IGF-2 increase in serum levels of people with breast cancer. But no relationship was found between IGF-1 and IGF-2 serum levels and age, and we suggest a large-scale study with more groupings. However, the results of our study indicate an association between IGF-1 and IGF-2 with breast cancer.

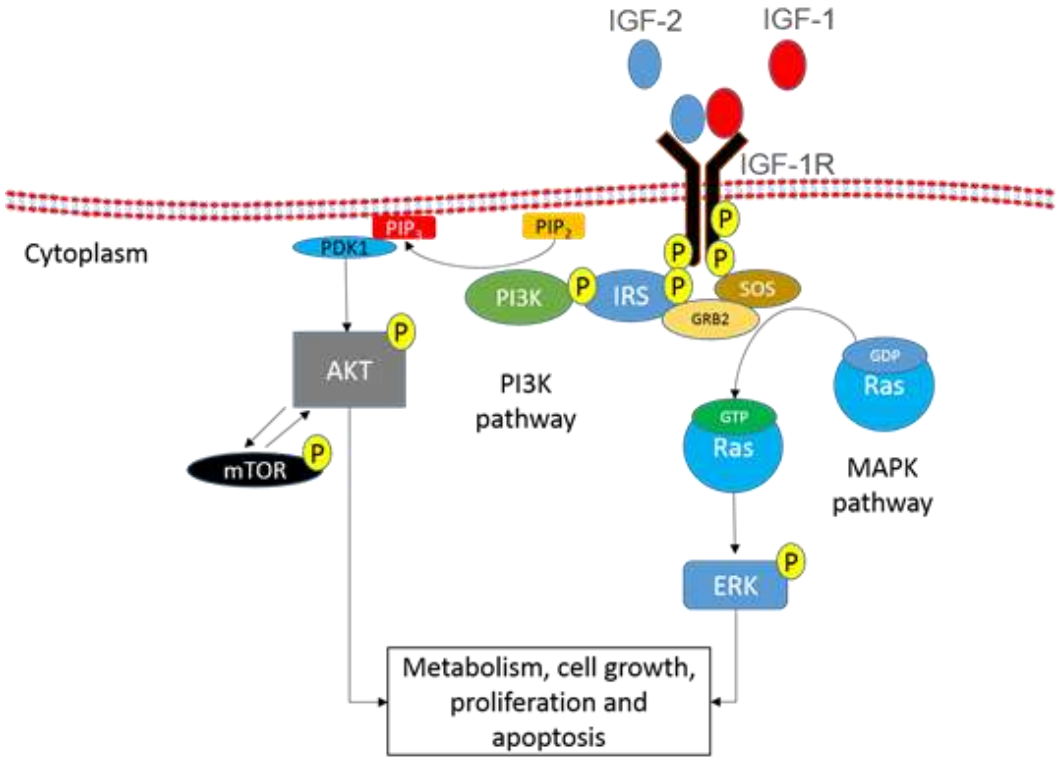

Figure 3. Simplified view of IGF-1 and IGF-2 signaling. IGF-1 and IGF-2 have effects on cell growth, metabolism, proliferation and apoptosis by activating the IGF-1 receptor.

Acknowledgments

This study was financially supported by Shahid Beheshti University of Medical Sciences (grant no.: 98532).

Ethics approval: This study was approved by the Ethics Committee of Shahid Beheshti University of Medical Sciences (IR.SBMU.MSP.REC.1399.246 (https://ethics.research.ac.ir/IR.SBMU.MSP.REC.1399.246)) with the commitment of the declaration of Helsinki.

Conflicts of Interest: The author(s) received no financial support for the research.

Authors' contribution: S.A, N.R: conceptualization, methodology, and investigation. N.R and M.T: supervision and project administration. S.A and M.M: collected samples and acquisition of data. S.A: formal analysis and visualization. S.A and N.R: writing and preparing the original draft. S.A, N.R, M.T and M.M: review & editing the manuscript. All authors approved the submitted version.

References

1. Khodadadi E, Ataei N, Mofid MR. The effect and mechanism of action of insulin-like growth factor-1 and insulin-like growth factor binding protein-3 in human breast cancer; A systematic review. J Isfahan Med Sch 2013; 31: 1560-7. (in Persian)
2. Feng X, Lin J, Xing S, Liu W, Zhang G. Higher IGFBP-1 to IGF-1 serum ratio predicts unfavourable survival in patients with nasopharyngeal carcinoma. BMC Cancer 2017; 17: 90.
3. Oh H, Eliassen AH, Beck AH, et al. Breast cancer risk factors in relation to estrogen receptor, progesterone receptor, insulin-like growth factor-1 receptor, and Ki67 expression in normal breast tissue. NPJ Breast Cancer 2017; 3: 39.
4. Key T, Appleby P, Barnes I, Reeves G; Endogenous Hormones and Breast Cancer Collaborative Group. Endogenous sex hormones and breast cancer in postmenopausal women: reanalysis of nine prospective studies. J Natl Cancer Inst 2002; 94: 606-16.
5. Espelund U, Cold S, Frystyk J, Ørskov H, Flyvbjerg A. Elevated free IGF2 levels in localized, early-stage breast cancer in women. Eur J Endocrinol 2008; 159: 595-601
6. Endogenous Hormones and Breast Cancer Collaborative Group; Key TJ, Appleby PN, Reeves GK, Roddam AW. Insulin-like growth factor 1 (IGF1), IGF binding protein 3 (IGFBP3), and breast cancer risk: pooled individual data analysis of 17 prospective studies. Lancet Oncol 2010; 11: 530-42.
7. Sedighi Pashaki A, Mohammadian K, Afshar S, et al. A randomized, controlled, parallel-group, trial on the

effects of melatonin on fatigue associated with breast cancer and its adjuvant treatments. Integr Cancer Ther 2021; 20: 1534735420988343.
8. Sedighi Pashaki A, Sheida F, Moaddab Shoar L, et al. A randomized, controlled, parallel-group, trial on the long-term effects of melatonin on fatigue associated with breast cancer and its adjuvant treatments. Integr Cancer Ther 2023; 22: 15347354231168624.
9. Livingstone C. IGF2 and cancer. Endocr Relat Cancer 2013; 20: R321-39.
10. Casa AJ, Dearth RK, Litzenburger BC, Lee AV, Cui X. The type I insulin-like growth factor receptor pathway: a key player in cancer therapeutic resistance. Front Biosci 2008; 13:3273-87.
11. Massarweh S, Osborne CK, Creighton CJ, et al. Tamoxifen resistance in breast tumors is driven by growth factor receptor signaling with repression of classic estrogen receptor genomic function. Cancer Res 2008; 68: 826-33.
12. Denduluri SK, Idowu O, Wang Z, et al. Insulin-like growth factor (IGF) signaling in tumorigenesis and the development of cancer drug resistance. Genes Dis 2015; 2: 13-25.
13. Brahmkhatri VP, Prasanna C, Atreya HS. Insulin-like growth factor system in cancer: novel targeted therapies. Biomed Res Int 2015; 2015: 538019.
14. Grimberg A, Cohen P. Role of insulin-like growth factors and their binding proteins in growth control and carcinogenesis. J Cell Physiol 2000; 183: 1-9.
15. Shanmugalingam T, Bosco C, Ridley AJ, Van Hemelrijck M. Is there a role for IGF-1 in the development of second primary cancers? Cancer Med 2016; 5: 3353-67.
16. Wu MS, Wang HP, Lin CC, et al. Loss of imprinting and overexpression of IGF2 gene in gastric adenocarcinoma. Cancer Lett 1997; 120: 9-14.
17. Bhusari S, Yang B, Kueck J, Huang W, Jarrard DF. Insulin-like growth factor-2 (IGF2) loss of imprinting marks a field defect within human prostates containing cancer. Prostate 2011; 71: 1621-30.
18. Rosendahl AH, Björner S, Ygland Rödström M, et al. Pre- and postoperative circulating IGF-I, IGFBP-3, and IGFBP-7 levels in relation to endocrine treatment and breast cancer recurrence: A nested case-control study. Front Oncol 2021; 11: 626058.
19. Werner H. Tumor suppressors govern insulin-like growth factor signaling pathways: implications in metabolism and cancer. Oncogene 2012; 31: 2703-14.
20. Christopoulos PF, Msaouel P, Koutsilieris M. The role of the insulin-like growth factor-1 system in breast cancer. Mol Cancer 2015; 14: 43.
21. Farabaugh SM, Boone DN, Lee AV. Role of IGF1R in breast cancer subtypes, stemness, and lineage differentiation. Front Endocrinol (Lausanne) 2015; 6: 59.
22. Osher E, Macaulay VM. Therapeutic Targeting of the IGF Axis. Cells. 2014; 8: 895.
23. LeRoith D, Holly JMP, Forbes BE. Insulin-like growth factors: Ligands, binding proteins, and receptors. Mol Metab 2021; 52: 101245.
24. Allard JB, Duan C. IGF-Binding proteins: Why do they exist and why are there so many? Front Endocrinol (Lausanne) 2018; 9: 117.
25. Griffeth RJ, Bianda V, Nef S. The emerging role of insulin-like growth factors in testis development and function. Basic Clin Androl 2014; 24: 12.
26. Yakar S, Adamo ML. Insulin-like growth factor 1 physiology: lessons from mouse models. Endocrinol Metab Clin North Am 2012; 41: 231-47, v.
27. Neirijnck Y, Papaioannou MD, Nef S. The insulin/IGF system in mammalian sexual development and reproduction. Int J Mol Sci 2019; 20: 4440.
28. Griffeth RJ, Bianda V, Nef S. The emerging role of insulin-like growth factors in testis development and function. Basic Clin Androl 2014; 24: 12.
29. De Meyts P. The insulin receptor and its signal transduction network. In: Feingold KR, Anawalt B, Blackman MR, et al., editors. Endotext. South Dartmouth (MA): MDText.com, Inc.; 2000-. Available from: https://www.ncbi.nlm.nih.gov/books/NBK378978/. Accessed Apr 16, 2016.
30. Tatulian SA. Structural dynamics of insulin receptor and transmembrane signaling. Biochemistry 2015; 54: 5523-32.
31. Hakuno F, Takahashi SI. IGF1 receptor signaling pathways. J Mol Endocrinol 2018; 61: T69-86.
32. Ha DP, Lee AS. Insulin-like growth factor 1-receptor signaling stimulates GRP78 expression through the PI3K/AKT/mTOR/ATF4 axis. Cell Signal 2020; 75: 109736.