Original Article

The Effect of Hypoxia-Inducible Factors Levels in Sera of Babylon Women with Breast Cancer

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Abstract

Background: The most common cause of death linked to advanced stages of breast cancer (BC) is metastases. Newly, biomarkers of tumors are important due to their probable use in the early detection of the tumor. Many types of cancer were associated with hypoxia. The action of the hypoxia-inducible factors (HIFs), the component of the response to hypoxia and its adaptive , improves cancer complications like immune suppression, disturbed metabolism, angiogenesis, invasive, and metastasis. HIFs became important aim in anticancer therapy designation. **Objectives:** The aim of the present education is to evaluate HIF 1α (HIF-1A) and HIF 2α (HIF-2A) levels in the plasma of Babylon women with BC and control groups. **Materials and Methods:** The case–control group was made up of 50 women (presumably healthy women), whereas the sick group was made up of 50 women with BC. In plasma, the concentrations of HIF-1α (HIF-1A) and HIF-2α (HIF-2A) levels were evaluated by enzyme-linked immunosorbent assay (ELISA), and the statistical analysis was conducted by the SPSS software. **Results:** The results showed elevated HIF-1A and HIF-2A levels in the patient's group compared with the control group ($P < 0.05$). **Conclusion:** The women with BC had elevated serum levels of HIF-1A and HIF-2A. Based on the results of this investigation, indicate the role of HIF-1A and HIF-2A as prognostic indicators in BC women.

Keywords: ELISA, hypoxia-inducible factor 1α and 2α, SPSS

Introduction

Breast cancer (BC) represents a heterogenous disease described as abnormal cell growth in the glands producing milk or in the ducts that provide milk to the breast nipples.[1] In breast tumors, the main used prognostic factors include: age, menstrual status, dimension of the tumor, status of node, and the stage of TNM. The serum markers were simple, and also have a significant role in a lot of types of cancer, yet there is uncertainty about their role in BCs. Generally, the arrangement of BC is into stages 0–4, dependent on the progression of the tumor stage 0: tumors of noninvasive status. Stages 1–3: tumors without distant metastasis. Stage 4: tumors with metastatic disease.[2]

Hypoxia accompanies numerous kinds of cancer. The hypoxia-inducible factor (HIF)-1 and HIF-2 isoforms display the same regular features, in different types of hypoxia they seemed active. BC has several distinctive methods of noncanonical inducible factors activity initiation hypoxia.[3]

HIFs are heterodimer proteins containing an oxygen-sensitive. The changes in gene expression of HIF produce changes in oxygen availability: in normal oxygen state, one of three HIF prolyl hydroxylases (PHD1 and PHD2) inserted an oxygen atom into a proline residue of HIF-1α (HIF-1A) or HIF-2α (HIF-2A) subunits, whereas hydroxylated subunits (HIF-α) bind with von Hippel–Lindau protein, for ubiquitination and proteasome degradation. Under hypoxia state, the hydroxylation inhibited and nonhydroxylated HIF-α subunits accumulated, for activation of transcription, HIF-1β dimerized and binding to HREs in target genes.[4] Factor

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inhibiting (FIH-1) modulated the transcription of HIF, by hydroxylation of asparagine residue, thus the binding of the coactivator proteins p300 and CREB binding protein (CBP) is blocked. Consequently, hydroxylation of HIF-α subunits by prolyl and asparaginyl regulates their half-life negatively and the activity of transcription, in an O_2-dependent manner.[5]

The deoxyribonucleic acid (DNA) is damaged by hypoxia, prompting mutation, breakdowns, oxidative base damages, and ultra-replication. Hypoxia causes transport and current of electrons disorders resulting in DNA damages, that induce the production of reactive oxygen species. Polyploidy also promoted by hypoxia, resulting from the replication of the genome without cell division. Most of the genes in cancer cells causing adaptation and metastasis.[6]

Cancer cells tend to be more malignant because hypoxia prefers resistant cells with more hypoxia. In addition, leads to resistance against radiation and cancer therapy; it can diminish the effect of photodynamic therapy. The vessels of blood sustenance the cancer cells growth until the tumor mass is more than 2 mm^3. Then, tumor growth is based on oxygen supplied by the growth of new blood vessels in angiogenesis. Yet, disorganized or leaky vessels formed by vascularization, lead to hypoxia or hypoxic areas.[7]

Materials and Methods

The current study involved 50 women (diagnosed with BC) from Al-Hillah. The samples of patients when coming to hospitals for checking and treatment, aged between 33 and 64 years during the period from December 7, 2022 to May 9, 2022 were taken. Patients group were separated into two stages and grades (II and IV), also into three subgroups according to ages (33–43), (44–54), and (55–64) years, and three other subdivisions rendering to body mass index (BMI; normal weight and overweight). The control group was composed of 50 women who appeared as healthy aged between 33 and 64 years. The ages were matched for healthy and patient groups. Mean ± SD was used for the expression of data. Student *t* test and the linear regression analysis were used for the analysis of data. The result stated an odd ratio, 95% confidence interval, and *P* value. The SPSS software (version 20) was used for statistical analyses. *P* value <0.05 () was considered significant.

Ethical approval

Before collecting samples for the study, the acceptance from the scientific committee in the Biochemistry Department of Babylon Medical College was taken. All the participants in the present study have an explanation of the aims and techniques to gain verbal approval.

Results

In the present study, HIF-1A and HIF-2A were considerably (P < 0.05) higher in breast cancer patients than in the control group, as indicated in (Table 1). In the second stage, HIF-1A rises and falls. When compared to the control group, HIF-1A levels in groups II and IV were higher (17.2 ± 0.7 and 9.8 ± 0.3, respectively), whereas HIF-2A increases in groups II and IV are 5.9 ± 0.4 and 8.2 ± 0.5, respectively. The results in (Tables 2 & 3) display an HIF-1A vs HIF-2A correlation significance of less than 0.05.

Discussion

In the present study, HIF-1A and HIF-2A significantly elevated (*P* < 0.05) in BC compared with the control group shown in Table 1. HIF-1A increases in stage 2 and reduces in stage 2, this finding is following another review like Kozal *et al.*[8] found that HIF-1is stabilized as a result of acute hypoxia, but this level reduces in elongated of hypoxia. An increase in the level of HIF-2 is observed in chronic hypoxia.

The conversion of HIF-1 to the HIF-2 isoform is regulated by numerous devices. First, HIF-2 isoform resisted hydroxylation by PHDs and FIH and other factors regulated it; then it displays stability greater than HIF-1.[9] The transcriptional activity of HIF-2 stimulates the hypoxia-associated factor and causes HIF-1 subunit ubiquitination and degradation. The second mechanism,

Table 1: Biochemical characteristics of the control and patient groups

Variables	Group	No	Mean ± SD	95% confidence interval for mean		Sig. *P* value
				Min	Max	
HIF-1A (ng/ml)	II	25	17.2±0.7	16.5	17.9	<0.05
	IV	25	9.8±0.3	9.6	10.1	
	Control	50	3.5±0.3	3.2	3.8	
HIF-2A (ng/ml)	II	25	5.9±0.4	5.5	6.3	<0.05
	IV	25	8.2±0.5	7.7	8.7	
	Control	50	2.2±0.1	2.1	2.3	

significant = *P* < 0.05

Table 2: A correlation (*r*) between the measured parameters against each other

Sequence	Variables against each other	Correlation (*r*)	Sig. *P* value
II	HIF-1A versus HIF-2A	0.62	<0.05
IV	HIF-1A versus HIF-2A	-0.32	<0.05

Table 3: Comparison between HIF-1A and HIF-2A levels according to BMI in women with breast cancer disease

BMI	HIF-1A	HIF-2A	*P* value
Normal weight	11.1±0.8	6.2±0.7	0.05
Obese	14.4±1.8	8.1±0.4	0.05

activated the expression of anti-sense RNA by HIF-2, alterations of chromatin, activation of the response element 1-silencing transcription factor, and microRNAs regulations. The isoforms of HIF-1 and HIF-2 display the same regulation appearances, they are active in diverse kinds of hypoxia and exposed to unlike or reverse effects.[10]

In physiology, the HIF participates in the development of the embryo, chondrogenesis, osteogenesis, circulatory system, adipogenesis, hematopoiesis, and immune system development. It shows a defensive effect in the disease of arteries, rejection of organ transplantation, wound healing, and colitis. In addition, the HIF factor causes sleep apnea pathogenesis and cancers.[11]

The association of regions with hypoxia and disturbance of HIF is unknown, signifying another mechanism of activation. Still, tumors from hematopoietic cells do not form tumors and stated upregulation of the hypoxia-inducible factor. In humans, about (88%) of normal tissues do not have HIF-1 because of rapid degradation. According to sample studies, in most normal breast tissue HIF-1 is expressed and shown higher level of HIF-1 in all samples of BC.[12] It is detected in most cancers, like prostate, lung, pancreas, and brain cancer. The HIF expression is detected in non-hypoxic cancer cells. The HIF-1 level correlated with the cell differentiation degree negatively, demonstrating that further undifferentiated and immature and special cells, like malignant cells, tend to have more HIF-1.[13] Diverse effects on metabolism by the activation of HIF, which is detected depending on the isoform. Similarly, HIF-1 and HIF-2 lower the pyruvate dehydrogenase kinase level, which causes additional oxidation. In addition, glutaminolysis involves the HIF-2 isoform, it produce adenosine triphosphate in reduced oxidative phosphorylation production or glycolysis. A variance is also seen in the genes of the factor. Though some genes overlap for both isoforms, they have distinctive aims. The activity of HIF-1 is to regulation of genes linked with glycolysis. It is supposed that some of the special aims of the HIF-1 are genes coding glucose transporter-1 (GLUT1), phosphoglycerate kinase (PGK), aldolase, mitochondrial hexokinase (HKII), and pyruvate kinase M2 (PKM2).[14]

Table 2, observed that HIF-1 positively correlated with HIF-2 in (stage 2) of BC and negatively correlated with HIF-2 in (stage 4) of BC. This finding disagrees with a study conducted by Bharti *et al*.,[15] no correlation was presented between HIF-1 and HIF-2, in BC. On the other hand, the level of factors-induced hypoxia correlates with a few characteristics of breast cancer like the type of molecule, receptors status, tumor size, age, and BMI as shown in Table 3.[16] HIF-1 was also found to be related to viral hepatitis.[17]

Conclusion

This study presents the significant difference between the HIF-1 and HIF-2 between BC and the control group. In addition variables like stage and BMI in women with BC disease affect the level of HIF-1 and HIF-2, which indicates the role of HIF-1 and HIF-2 as prognostic markers in women with BC disease.

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Conflicts of interest

There are no conflicts of interest.

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