

Research paper

Study of blood groups antigens frequency for ductal breast carcinoma patients in some Iraqi women

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ABSTRACT

100 blood samples collected from patients with ductal breast carcinoma for some women in Iraq. Blood groups antigens frequency (ABO, Lewis, MNS, and Lutheran) is measured using Hemagglutination test. The results showed that no statistical significant for two blood types (A, O) in ductal breast carcinoma patients and control groups. Whereas, blood type (B) in patients is lowest (10%) than in control groups (24%), and there is statistical significant of $p<0.01$. The Lewis blood groups in patients have different frequency compare with control groups. Where, Le ($a^- b^+$) blood groups in patients is lowest (40%) than in control groups (55%) with statistical significant ($p<0.05$), Le ($a^+ b^-$) blood groups in patients are lowest (29%) than in control groups (31%) with no statistical significant, Le ($a^- b^-$) blood groups in patients is highest (23%) than in control groups (10%), and Le ($a^+ b^+$) blood groups in patients is highest (8%) than in control groups (4%) with statistical significant ($p<0.05$). Whereas, MNS groups in patients are highest (77%) than in control groups (57%) with statistical significant ($p<0.01$), Lutheran ($a^- b^-$) groups in patients are highest (31%) than in control groups (6%) with statistical significant ($p<0.01$) and Lutheran ($a^- b^+$) groups in patients are lowest (65%) than in control groups (83%) with statistical significant ($p<0.01$). Finally, the results of the present study indicate that there is a correlation between the presence of molecules responsible for blood groups and the presence of ductal breast cancer as select this interdependence depending on the presence in the patient groups and it different in control groups.

Keywords: ABO, Lewis, MNS, Lutheran, Ductal Brest cancer

Introduction

Lies the importance of the major blood group (ABO) and a group of other blood transfusion such as Lewis, MNSs, Lutheran, Kidd, Duffy and Kell, as antigens in the process of mismatches in blood transfusions. After the discovery of the blood group (ABO) by Landsteiner in 1900, studies have focused on expanding the knowledge of these molecules being Adhesion molecules in parasitic and viral or bacterial infections (Garratty, 2005), they also play a key role like associated molecules of cancer (Seham *et al.*, 2008). In addition, they are associated with many other diseases. (Anstee, 2010). Studies have indicated to the importance of blood group (Lewis) after discovered by Mourant, 1946 because of the complexity of the installation, which cause a change in cell surface markers and this group includes many of the patterns as well as enter in manufactured it the enzymes carrier of fayyokoz and galactose sugars and these enzymes controlled by several genes. Many studies have also indicated to the existence of a relationship between antigens and antibodies of this group with the incidence of many diseases such as cancer, heart and kidney (Wazirali *et al.*, 2005; Holt *et al.*, 2004; Torrado *et al.*, 2000). This group plays a role of adhesion factors and Metastases of cancer cells (Takada*, et al*., 1991). In addition, the genes controlling on the construction of this group have an important role for many other diseases. The aim of this study is find blood groups frequency (ABO, Lewis, MNS, and Lutheran) in healthy and patients groups.

Materials and Methods

100 blood samples collected from women patients with ductal breast carcinoma from radiation and nuclear medicine hospital, Iraq. These patients have been diagnosed by specialist doctors by cellular examination by taking a sample of breast tissue by Fine Needle Aspiration (FNA) method, as it was determined the cancer cells of the breast tissue by microscopic and depending on the cellular changes to breast tissue. Where, treated women with chemotherapy or radiation are excepted, as well as pregnant patients are excepted and so the disappearance of the Lewis blood group antigens at this group of patients. 100 blood samples from women of healthy are collected from the National Center for Blood Transfusion and Teaching Baghdad Hospital, Ministry of Health. Blood samples from the patients of 5 ml (3 ml in plastic test tube and 2 ml in tube with 5-ethylenediamine tetra acetic acid (EDTA)) were collected using syringes. The samples (3 ml in plastic tube) are left for 30 minutes at room temperature where it was clotting. Tube of blood samples is placed in a centrifuge at speed (3000) rpm for 10 minutes and then blood serum was isolated. Finally, blood serum samples are divided into small quantities in eppendorf tubes and stored at a temperature - 40 ° C until use. The samples (2 ml in tube with EDTA) are used for blood grouping.

Stereotyping blood groups

a) Stereotyping ABO blood group by laboratory tubes method

Put a drop of the Anti-A in the test tube, as well as put a drop of the Anti-B in another test tube and each tube was added one drop of blood cells prepared in this study. The tubes were left at room temperature for 5 minutes. Where it was put to a

centrifuge for 30 seconds and at speeds of 3000 r / min and then mix the contents of each tube gently and recorded the results of positive and negative agglutinations.

b) Stereotyping of Lewis blood groups by laboratory tubes method

Put a drop of the Anti-Lea in the test tube, as well as put a drop of the Anti-Leb in another test tube and each tube was added one drop of blood cells prepared in this study. The tubes were left at room temperature for 15 minutes. Where it was put to a centrifuge for 30 seconds and at speeds of 3000 r / min and then mix the contents of each tube gently and recorded the results of positive and negative agglutinations.

c) Stereotyping of blood groups (MNSs) by laboratory tubes method

Put a drop of the Anti-M, Anti-N, Anti-S, and Anti-s in the four test tubes, each tube was added one drop of blood cells prepared in this study. The tubes were left at room temperature for 30 minutes. Where it was put to a centrifuge for 30 seconds and at speeds of 3000 r / min and then mix the contents of each tube gently and recorded the results of positive and negative agglutinations. Tube containing the cells and antibody stuck Anti-s was placed in the incubator at a temperature 37 ° C for 30 minutes, then wash stuck three times with saline solution and using centrifugal fast 3000 r / min for two minutes. Two drops of Anti human globulin (AHG) added to blood cells sediment. Tube placed to a centrifuge for 30 seconds and at speeds of 3000 r / min and then mix the contents of each tube gently and recorded the results of the positive and negative agglutinations.

d) Stereotyping of Lutheran blood groups by laboratory tubes method

Put a drop of the Anti-Lua in the test tube, as well as put a drop of the Anti-Lub in another test tube and each tube was added one drop of blood cells prepared in this study. The tubes were left at temperature of 37 ° C for 60 minutes, then wash stuck three times with saline solution and using centrifugal fast 3000 r / min for one minutes. Two drops of Anti human globulin (AHG) added to blood cells sediment. Tube placed to a centrifuge for 30 seconds and at speeds of 3000 r / min and then mix the contents of each tube gently and recorded the results of the positive and negative agglutinations. Statistical analyses of data were conducted using SPSS software (2012) for Windows. Equality of several means was tested using one-way classification. The data were analyzed using Qi-square test. Various statistical analyses were performed for the data. Many published statistical analyses quote p-values as ≥ 0.05 (no significant), < 0.05 (significant), and < 0.01 (highly significant) (Almayahi, 2013; Laith et al., 2016).

Results and Discussion

The results showed that frequency of blood type (A) found to be 25% and 27% in patients and healthy, respectively as shown in Table 1. The statistical analysis showed that there are no statistical significant between two frequencies. Table 1 showed that frequency of blood type (B) is lowest in patients (10%) than in healthy (24%) ($X^2 = 6.814$, $p < 0.01$), frequency of blood type (O) is highest in patients (43%) than in healthy (38%) and no statistical significant between two frequencies, and frequency of blood type (AB) is lowest in patients (22%) than in healthy (11%). The statistical analysis showed that there are statistical significant between patients and healthy ($X^2 = 3.861$, $p < 0.05$). Blood type frequency (AB) is high and blood type frequency (B) is low, this may be because convert blood type group (B) to AB group by convert phenomena of blood group. **(Table 1)**

Al-Musawi, 2007 showed there are no statistical significant between patients and healthy woman regard to blood groups of A and B, this result is agreed with this study, whereas this study not agree with Al-Musawi, 2007 regard to blood groups of AB and B. Costantini *et al*., (1990) showed that there are no relationship between ABO blood groups and breast cancer. Tryggvadottir, 1988 studies breast cancer patients and found that there are statistical significant in blood groups (B). Akammu *et al*., 2002 concluded that there are no limit relationship between ABO blood groups and breast cancer. It concluded that the results are different in literature review and this difference may be because sample size. Lewis blood groups in patients have different frequency compare with control groups as shown in (**Table 2)** Where, Le ($a^- b^+$) blood groups in patients is lowest (40%) than in control groups (55%) with statistical significant ($X^2 = 3.496$, $p<0.05$), Le ($a^+ b^-$) blood groups in patients is lowest (29%) than in control groups (31%) with no statistical significant, Le ($a^- b^-$) blood groups in patients is highest (23%) than in control groups (10%) at $X^2 = 2.868$ with statistical significant ($p<0.05$), and Le ($a^+ b^+$) blood groups in patients is highest (8%) than in control groups (4%) at $X^2 = 2.673$ with statistical significant ($p<0.05$). When, Le ($a^+ b^-$) increased then Le ($a^- b^-$) decreased, this mean convert Le ($a^+ b^-$) to Le ($a^- b^-$).**(Table 2)**

Al-Musawi, 2007 showed there are statistical significant between patients and healthy woman regard to Le($a^+ b^-$) blood groups, this result is no agree with this study. Idikio and Manickavel, 1993 showed that Lewis blood groups are low in gene expression and also loss of Le^a, Le^b, and H from breast cancer tissue. Tohru et al., 1998 showed loss of Lewis antigens from breast cancer tissue and lymph nodes under the armpit, this mean increased in Le (a- b-) group frequency. As the this group are made in the tissue and then move to the plasma to stick to the red blood cells, any defect in the expression of antigens in the tissues of this group reflects on what is present in the blood cells. This result is agreed with this study. Steplewska-Mazur, et al. 2000 showed there are increased in Le^a antigens in breast cancer tissue. MM genotype frequency groups in patients are lowest (11%) than in control groups (29%) with statistical significant ($X^2 = 6.358$, $p<0.001$). Whereas, MNS groups in patients are highest (77%) than in control groups (57%) with statistical significant ($X^2 = 6.944$, $p<0.001$), and NN genotype frequency groups in patients are lowest (12%) than in control groups (14%) with no statistical significant as shown in **(Table 3)** SS genotype frequency groups in patients are highest (28%) than in control groups (15%) with statistical significant ($X^2 = 4.852$, $p<0.05$), Ss genotype frequency groups in patients are lowest (30%) than in control groups (36%) with no statistical significant, ss genotype frequency groups in patients are lowest (31%) than in control groups (43%) with statistical significant ($X^2 = 4.283$, $p<0.05$), $S^- s^-$ genotype frequency groups in patients are highest (11%) than in control groups (6%) with statistical significant ($X^2 = 2.639$, $p<0.05$), and as shown in **(Table 3)**.

Table 1: Blood group frequency (ABO) in patients and healthy woman.

	Patients			Controls			
Blood Group	**Number**	**Total**	**%**	**Number**	**Total**	**%**	**Qi-square**
A	25	100	25	27	100	27	0.263
B	10		10	24		24	** 6.814
O	43		43	38		38	1.095
AB	22		22	11		11	*3.861
Qi-square	……	……	**6.219	……	……	**6.219	……

**p<0.01
* p<0.05

Table 2: Lewis blood group frequency in patients and healthy woman.

	Patients			Controls			
Blood Group	**Number**	**Total**	**%**	**Number**	**Total**	**%**	**Qi-square**
Le (a^+ b^-)	29	100	29	31	100	31	0.538
Le (a^- b^+)	40		40	55		55	*3.496
Le (a^- b^-)	23		23	10		10	*2.868
Le (a^+ b^+)	8		8	4		4	*2.673
Qi-square	……	……	**7.390	……	……	**10.619	……

**p<0.01
*p<0.05

Table 3: MNS blood group frequency in patients and healthy woman.

	Patients			Controls			
Blood Group	**Number**	**Total**	**%**	**Number**	**Total**	**%**	**Qi-square**
MM	11	100	11	29	100	29	**6.358
MN	77		77	57		57	**6.944
NN	12		12	14		14	0.507
SS	28	100	28	15	100	15	*4.852
Ss	30		30	36		36	1.266
ss	31		31	43		43	*4.283
S-s-	11		11	6		6	*2.639
Qi-square	……	……	8.417**	……	……	**6.831	……

**p<0.01
*p<0.05

The MNS genetic antigens are works like receptors in cellular adhesion dynamics **(**cytokines**)** (Hassoun *et al*., 1998). Studies showed that there is a relationship between the incidence of asthma in children and the pattern (M^+N^-) (Bottini *et al*., 2005). Al-Musawi, 2007 showed there are no statistical significant between patients and healthy woman regard to MM and MN blood groups, this result is no agree with this study. Several studies have pointed to the role of chains (MN, Ss) such as antigens associated with tumors. Otsuka et al., 1991 showed change in the composition of T antigen, which is the base material to form part of a series of diabetes MN in the breast tissue of women with malignant tumors, compared to women with a tumor is not malignant. In addition to, the role of antigen-N in the spread of tumor cells to cancer of the liver and pancreas. Phipps and Perry, 1989 showed that there are statistical significant for SS genotype frequency groups between patients and healthy. In this regard has to be considered with interest to accompany the rise of this group of MNS or those with breast cancer or lack of it in of healthy women has been so linked to the role of microorganisms from viruses, bacteria, parasites, and others in the establishment of the disease, so that the MNS molecules establish a tentative receptor-ligand link between host and microorganisms. Relationship of MNS group in invading microorganisms tissue. Rygiel et al., 1985 showed

Table 4: Lutheran blood group frequency in patients and healthy woman.

Blood Group	Patients			Controls			
	Number	Total	%	Number	Total	%	Qi-square
Lu ($a^+ b^-$)	3	100	3	11	100	11	*2.947
Lu ($a^- b^+$)	65		65	83		83	**6.395
Lu ($a^- b^-$)	31		31	6		6	**6.587
Lu ($a^+ b^+$)	1		1	0		0	0.003
Qi-square	……	……	**11.703	……	……	**14.240	……

**p<0.01
*p<0.05

O-linked bond in the chain Ss contribute to facilitating the process of invading blood cells by the plasmodium falciparum parasite. Calhoun, 1999 noted low invasion of blood cells by the parasite in people living with Mk gene that encodes for the $M^-N^-S^-s^-$ style losses for chains MN and Ss. Lutheran ($a^- b^-$) groups in patients are highest (31%) than in control groups (6%) with statistical significant ($X^2 = 6.587$,$p<0.01$), Lutheran ($a^- b^+$) groups in patients are lowest (65%) than in control groups (83%) with statistical significant ($X^2 = 6.395$, $p<0.01$), Lutheran ($a^+ b^-$) groups in patients are lowest (3%) than in control groups (11%) with statistical significant ($X^2 = 2.947$, $p<0.05$), and Lutheran ($a^+ b^+$) groups in patients are highest (1%) than in control groups (0%) with no statistical significant as shown in **(Table 4)**. Lutheran blood group plays an important role in coating cell link operations in the basal lamina and this group belongs to what is known as basal cell adhesion molecules (B-CAM). These molecules have an important role in cell adhesion. As for the relationship recorded a rise genotype Lu ($a^- b^-$) in the patients can be associated with the dialectical relationship between adhesions and loose from their cells in cancer cases. **(Table 4)**

Conclusions

In this study showing that there are different effects of the overlap between the different blood groups and the sickest cancer. With regard to MNSs blood groups, it has been observed high MN genotype frequency and low MM genotype frequency ($p <0.001$) in the patients. It observed significant increase in the genotype frequency Lu (a-b-) for ductal breast cancer patients.

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