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Research Paper / Article / Review

USING OF SOME IMMUNOLOGICAL AND HEMATOLOGICAL PARAMETERS IN WOMEN INFECTED WITH TRICHOMONAS VAGINALIS

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Abstract: The study was conducted on 72 women and 44 infected with T. vaginalis, and 28 healthy women in Zakho hospitals during the period from January to October, 2017. From infected and healthy women vaginal swab was carefully collected and five ml was the total blood collected from each clinical suspected woman with T.vaginalis infection and non-suspected women(as control group).used two sterile swab s, first swab used for direct microscopic examination, while the second swab was used for some trichomonas rapid test, then quantify specific cytokine concentration (IL-2, IL-8, and CRP) The infection with T.vaginalis in clinical suspected women determined by using the wet amount microscope, ratio and infected women numbers by wet mount. The aim of the study was to characterize the systemic immune response in women with trichomonas's as compared with uninfected women. The total sera level of IL-2, IL-8 and CRP showed significant difference and Showed significant decrease of HB, PCV, MCV, MCH and MCHC as well as was increase of level lymphocyte, monocyte, neutrophil and basophil in infected woman's compared to healthy woman's.

Keywords: T. Vaginalis, Immunological Parameters, Cytokines, Hematological Parameter.

1. INTRODUCTION :

Trichomonas vaginalis It is one of the flagellated protozoan parasite that causes Trichomoniasis by affecting the urinary genital tract in humans and is one of the most important causes of non-viral genitourinary sexually transmitted infection (STI) with a prevalence rate of 180 million annually according to WHO estimates (1) T. vaginalis is the most widely studied parasite of all the trichomonas. This urogenital pathogen varies in size and shape, with the average length and width being 10 and 7 μ m, respectively (2). Physiochemical conditions do alter the appearance of the parasite. In axenic culture, the shape of the protozoan tends to be more uniform, i.e., pear shaped or oval (3), but the parasite takes on a more amoeboid appearance when attached to vaginal epithelial cells (3,4). T. vaginalis has only trophozoite stage with a size of (7-32 µm) and is pear shaped. It possesses 5 flagellates, 4 of which are forward with a fifth flagella that bends back along the undulating membrane and lacks the cyst stage and possesses a central nucleus and axostyle, Trophozoite is divided by binary fission. The parasite is an aero tolerant anaerobe that dissolves whole carbohydrates into short chains of organic acids Especially (acetate and lactate) Regardless of the presence of oxygen. The parasite produces hydrogen molecules in the absence of oxygen, and this reaction occurs in hydrogenases that like mitochondria (which are not present) in T. vaginalis, which are energy producing organelles surrounded by two membranes (5). Trichomoniasis is the most prevalent non-viral sexually transmitted infection in the world (6) Trichomonas vaginalis, the causative agent is a protozoan parasite infecting the urogenital tract of both females and males (7). and Trichomoniasis accounts to almost half of curable sexually transmitted infections according to the world health organization (8,9) Women with trichomoniasis may be asymptomatic or may experience various symptoms, including vaginal discharge and vulvar irritation. Men with trichomoniasis may experience no gonococcal urethritis but are frequently asymptomatic. (10)



The general infection areas include urethra, external genitalia, prostate, and epididymis.(11) It is also a known leading cause of prostatitis and urethritis.(12) Furthermore, serious complications may be associated with trichomoniasis such as premature rupture of the placental membranes, premature labor, and low birth weight in pregnant women infection, infertility, cervical cancer, pelvic inflammatory disease, birth outcomes, and increase in human predisposition to immunodeficiency virus (HIV), its transmission, and acquisition.(13,14) Infection with *T. vaginalis* is associated with higher genital HIV-1 levels. The treatment of women infected with *T. vaginalis* results in a 4.2-fold reduction in the quantity of HIV-1 in vaginal secretions. (13) Some recent studies have shown that trichomoniasis may be considered as one of the predisposing factors for some human disease such as prostate and cervical cancer. (15) The outcome of infection with *Trichomonas* may be due to genetic variability of the isolates and the host immune response. (16).

Laboratory diagnosis of T. vaginalis infection is based on their covey of the motile organism in the vaginal discharge or mucosal scraping of the vagina in female and urethral discharge in male, and it is able to detect the protozoa at concentration as low as three organisms (17). The traditional clinical diagnosis of vaginal infection is based on a history of patient, clinical finding observed during the vaginal specimen. This last one provides the most objective information. A microscope wet amount examination of the specimen permits, detection of the motile protozoa T. vaginalis (18). Although T. vaginalis is the most common cause of non-viral STD (19).

The present study was aimed to assess some immunological parameters by assess serum concentrations of cytokines, (IL2, IL-8 and (TNF)-alpha) and (CRP) were compared between infected and uninfected women, also study of blood parameter (WBC, PCV, HB, MCV, MCH, MCHC) in infected women and uninfected with Trichomonas virginals.

2. MATERIALS AND METHODS :

The study was conducted on 72 women and 44 infected with T. vaginalis, and 28 healthy women in Zakho hospitals during the period from January to October, 2017. From infected and healthy women vaginal swab was carefully collected from the posterior vaginal fornix after putting the patient at a lithotomic position and taking swab after opening the vagina by a sterile speculum, the swab are immersed in a tube with1 ml of a sterile normal saline ,and five ml was the total blood collected from each clinical suspected woman with T.vaginalis infection and non-suspected women(as control group) by disposable syringe, the blood samples were drawn in sterile plain tubes and remains for 30 minutes at room temperature. After that the samples were centrifugation at 3000 rpm for 5 minutes to separate the serum and collected in other sterile tubes, each sample of serum was divided into four parts; each of them was kept in deep freeze at -20C°. The samples were collected during vaginal examination , vaginal swab was collected from patients using two sterile swab s, first swab used for direct microscopic examination , this swab mixed with drops of normal saline and placed on a slid and examined at (X40), Positive results were defined as the presence of one or more Trichomonas with characteristic motility jerky movement and morphology while the second swab was used for trichomonas rapid test: assay In order to quantify specific cytokine concentration (IL-2, IL8) in the patient's serum, ELISA kit was used.

The swab was examined in wet mount preparation and five ml was the total blood collected from each clinical suspected woman with T. vaginalis infection and non-suspected women (as control group) by disposable syringe. Four ml from each of blood samples were drawn in sterile plain tubes and remains for 30 minutes at room temperature. After that the samples were centrifugation at 3000 rpm for 5 minutes to separate the serum and collected in other sterile tubes, each sample of serum was divided into three parts; each of them was kept in deep freeze at -20C°. The remaining one ml of blood was drawn in tube with anti-coagulated EDTA which was used for determination the hematological parameters.

Statistical analysis was using the computer software and SPSS system, and take Percentage as statistical significance.

3. RESULTS :

The study was conducted on 72 women, 44 of them infected with T. vaginalis, and 28 healthy women in Zakho hospitals during the period from January to October, 2017. detected by strip test. The total level of IL-8 in serum showed significant, difference between Woman infected (93.54) and Healthy woman .(78.33) However, the total sera level of IL-2, showed significant difference between Woman infected and Healthy woman respectively 97.66 and 45.43 as well



as the total sera level of CRP showed significant difference between Woman infected 1.03 and Healthy woman 0.356 in table (1).

Table 1: Compare between Healthy woman and infected woman by assess some immunological parameters.

Group	No.	Serum concentrations of cytokines		
		IL-2	IL-8	CRP
Healthy woman	28	45.43	78.33	0.356
Woman infected	44	97.66	93.54	1.03
LSD value		15.534	15.532	0.148

The results have shown a significant decrease in hemoglobin concentration, packed cell, volume, mean corpuscular volume, mean corpuscular hemoglobin concentration in infected woman's with T. vaginalis compared to the healthy woman's, as seen in table (2). Differential Leukocyte Percentage % in infected woman's suffering from T. vaginalis infection exhibited a significant increase as compared to the healthy woman's The result of differential type of lymphocyte, monocyte, neutrophil and basophil in infected woman's shown that the significant increase compared to healthy woman's but the eosinophil showing non-significant change in infected woman's compared to healthy woman.

Table (2): Hematological parameters between Healthy woman and infected woman and differential leukocyte Percentage for woman suffering from T. vaginalis infection.

Hematological parameters	Healthy woman $(n = 28)$	T.vaginalis woman (n = 44)
Hb	11.043	8.755
PCV	32.555	29.766
MCV	81.546	70.056
MCH	23.278	22.211
MCHC	26.822	24.066
Neu.	51.211	53.777
Lym.	22.056	25.878
Mon.	6.043	8.012
Eos.	2.107	2.125
Bas.	2.032	2.113

4. DISCUSSION :

Sexually transmitted diseases are a major global cause of acute illness, infertility, long term disability and death, with severe medical and psychological consequences for millions of men, women and children (20), may be due to the fact that there is no safe and effective method of prevention of trichomoniasis. Because of the potential side effects and clinical failures associated with the therapy drug of choice, metronidazole and the reemergence of resistant strains novel strategies are needed for the control of T. vaginalis infection, including vaccine production (21). The cytokine responses in various microbial diseases have been reported with varying observations, depending upon distinct patterns of immune responses stimulated by different microbes and unique mechanisms for evading specific immunity (22). Data from the present study showed highly significant increase in the concentration of IL-2 in serum of patients infected with T. vaginalis in comparison with control group., suggesting that Cytokines and chemokines provide a mechanism for initiation, amplification or containment of inflammation during disease status. Data from the present study showed increase in the concentration of IL-8 in serum and significant increase in serum of patients infected with T. vaginalis in comparison with control group. This is agreement with the study has shown that T. vaginalis induces blood monocytes to produce large amounts of bioactive IL-8, mainly by membrane components of T. vaginalis (MTV). Monocyte-derived IL-8 induced by MTV was dose and time dependent (23). The elevation of CRP in the serum of infected women with trichomoniasis in this study represents a biologically plausible link between a local vaginal infection and a systemic response. CRP is an acute-phase protein produced in response to cytokine stimulation and play is a role in a variety of responses to infectious agents. CRP is a known marker of systemic inflammation. Because labor is a systemic process,



it may be that there is systemic inflammation in the pathway leading to birth both at term and preterm (24). The presence of increased C-reactive protein in the sera of T. vaginalis infected women suggests that the impact of the immunoinflammatory reaction to the parasite exceeds the boundaries of the re productive tract mucosa (25).

The present results have revealed a significant decrease in concentration of Hb, PCV in infected woman s with T. vaginalis infection compared to healthy woman. This result may be due to hemolysis of RBCs and phagocytosis by T. vaginalis parasite and the increased bleeding through menstrual cycle and this leads to decrease in the number of RBCs and the hemolysis of RBCs may lead to decrease in the Hb concentration (10;11). One of the most frequent causes of anemia in T. vaginalis infection The results have shown a decrease in MCV, MCH and MCHC in women infection with T. vaginalis parasite compared to healthy woman's. A decrease in MCV may be due to a decrease in Hb and decrease in MCHC is caused by iron deficiency anemia that lead to decrease in formation of Hb in RBCs. The results also provided a decrease in level of PCV in patient women compared to control group; this result may be due to a decrease in RBCs counts or due to decrease in MCV caused by decreased level of Hb in RBCs. About 50% of infected women with T. vaginalis punctuate hemorrhages can be observed (2). The data of this study indicated a significant increase in WBCs; these due to an increase in the number of monocyte, lymphocyte, neutrophils, and Basophils because the infection with this parasite causes stimulation immune system of host humeral and cellular. The results also revealed Basophilia associated with patients who suffering from T. vaginalis infection; these phenomena are not fully understood. The reason for this observation may be attributed to allergy disorder which is one of symptoms of T. vaginalis infection these allergy causes.

5. CONCLUSION :

This present study there is significant increase in IL-2, IL-8 and CRP concentrations in the serum of women infected with T. vaginalis in comparison with the healthy women. This indicates a stimulation of the humoral immune response during the infection with T. vaginalis. and Showed significant decrease of HB, PCV, MCV, MCH and MCHC A decrease in MCV may be due to a decrease in Hb and decrease in MCHC is caused by iron deficiency anemia that lead to decrease in formation of Hb in RBCs The results also provided a decrease in level of PCV and the hemolysis of RBCs may lead to decrease in the Hb concentration as well as was increase of level lymphocyte, monocyte, neutrophil and basophil but the eosinophil showing non-significant change in infected woman's compared to healthy woman's. because the infection with this parasite causes stimulation immune system of host humeral and cellular, as well as allergy disorder is one of symptoms of T. vaginalis infection.

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