Expression of Pro Inflammatory Cytokines and Anti-Chlamydia Antibody in Bacterial Ocular Infection.

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Abstract.

Current study was done to investigate the bacterial causes of eye infection for 130 patients with different ages in Babylon city , the study includes swab for eye and sample of blood. Bacteriological diagnostic for eye swab which taken from different site include Conjunctivitis, Dacryocystitis, Lidinfection showed variation in types and number of isolates . Gram positive bacterial isolates include *Staphylococcus aureus* 67 (38.06%), Which represent the predominating bacteria, *Staphylococcus epidermidis* 18 (10.22%), While gram negative bacteria represent *Escherichia coli* 40 (22.7%), *Neisseriae gonorrhoeae* 5 (2.84%).

Detection of *Chlamydia trachomatis* reveals only one positive case out of 42 eye infected case for anti Chlamydia IgG –IgM (3.7 %),

Immunological test of Cytokine IL-6 in patient show increased in their concentration compared with healthy at P< 0.05 reached 8.52 pg/ml especially at age group 41-50.

IFN-γ also show increased in their concentration in patient than controls although, there is no significant differences between patient and control groups. .

Key words ; IFN gamma , IL-6 , eye infection , bacterial isolates ,*Chlamydia trachomatis*

Introduction

Exposed the human eye, like other members of the body, to the effects of so-called eye irritation that leds to various miserable including inflammation and other disorders (Mendell, 1993). There are irritants to the eye caused by personal behavior for some people itchy skin and exposure to sand, use of contact lenses and make-up (Wolkoff et al., 2003). Microorganisms represents the major causative agents of eye infection, so the rapid diagnosis and identification of these microorganisms is a crucial tools for protection and treatment (Todar ,2011).

Studies   have   shown   that   most   bacterial   causes   of   eye   infections *Staphylococcus aureus, Staphylococcus* epidermidis, *Streptococcus Pneumonia* in addition to others   (Hass et al., 2012; Seal et al., 1982).Local and systemic host defenses play important role in reducing an injury, eye are also had several immunological defenses against physical distressed . These barriers includes , eyelashes eyelid cilia which operates filtration molecules in order to prevent contamination entering the optical surface - and that the process of opening and closing the eye-hand arises when the stick foreign objects and motivated the immune system in the case of injuries occurring to the eye caused by the presence of bacteria or other neighborhoods (Ramesh et al ,2010)

  Body defenses in eye components such as that of lacimal glands and conjunctiva acts as a defensive through secretion of specific types of Immunoglobulins and other immune molecules that have a distinct role in cellular differentiation and growth (Rus et al ,2005). IL-6 is produced by different types of cells during various injuries such as trauma and microbial infections , the elevation in its concentration associated with medical conditions and could work as a Pro-and anti inflammatory cytokine (Jones et al., 2001).

The IL-6 is an important factor in the host defenses mechanical and plays an important role in several diseases with various injuries and the high level of IL-6 possible discovery serum found in body fluids depending on the location of injury was found high level in the vitreous of the eye is longer broker infections common (Hoekzema et al., 1990). Interferon IFNs are one group of Cytokines that play an important role in the immune response they includes IFNγ, IFNβ, IFNα, and they have a role against the viral infection through the activation of interferon-receptor (Diao et al ,2012). Previous studies pointed that IFNγ production was inducing with allergic inflammation of allergic conjunctivitis and diminished it (Fukushima et al , 2006).

Materials and methods

Five ml of blood were withdrawn from each patients and control placed in plain tube , let in room temperature for 30 minutes for coagulation , centrifuged at 3000 rpm for serum separation .The collected sera were stored at -20 °C until used in immunological investigations including Elisa assay for detection of IL6 and IFN γ concentration in patients sera according to company instructions   (Bioscience).

Searching for Chlamydia *trachomatis* were carried out depending on serological detection of anti-Chlamydia IgM-IgG in sera of eye infected infants according to company instructions (Vircell , Spain)

Transport sterile cotton swabs and different cultural media were used for cultivation and isolation of bacteria from different eye sites (Baron *et al.* , 1994). The bacterial isolation and identification was performed according to the recommended diagnostic procedures ,the final identification was performed biochemically by using different biochemical reagents (MacFaddin ,2000) .

Results

Bacteriological investigations for eye swabs obtained from eye infected patients show variation in number of bacterial isolates with different eye locations , Staphylococcus aureus reveals high percentage of isolates , while Neisseria gonorrhea represented the lower percentage of these isolates . Furthermore ,lidinfected patient swabs show more susceptibility to bacterial infection than other eye loci (table 1).

Table (1) show type of bacteria which isolate from patient infected with eye infection on different type.

Type of bacteria	Conjunctivitis	Dacryocystitis	Lidinfection	Total number
S. aureus	28 46%	12 (20%)	27(46.5)	67(38.06%)
S. epidermidis	6 10%	0	12 (20%)	18(10.22%)
E. coli	8(13.3%)	22(37.93%)	10(17.2%)	40(22.7%)
N. gonorrhoeae	2(3.33%)	1(1.7%)	2(3.44%)	5(2.84%)

.Serological test reveals only one positive case out of 42 eye infected case for anti Chlamydia IgG –IgM (3.7 %), this positive case appear with less than six month age infant (table 2).

Table (2) . Serological identification for anti-*Chlamydia trachomatis* IgG –IgM.

Age period	Samples	male	female	+ve results	%
< 6 months	27	19	8	1	3.7%
7months-1year	3	1	2	0	0%
1-5 years	12	6	6	0	0%

Immunological investigations for eye infected patients pointed out inducing of immune response through elevation in some interleukin concentrations in their sera .

IL-6 concentrations reveals elevation in all patients sera compared with healthy persons (table 3).

Table (3) show values of IL-6 in patient infected with eye infection.

Age group	IL-6 pg/ml (infected)M±SD	IL-6 pg/ml (control)M±SD
10-2	1.72±5.28	1.73±0.43
20-11	0.17±2.78	2.16±0.12
30-21	1.76±5.06	2.4±0.1
40-31	2.42±0.08	2±0.1
50-41	±8.52±2.56	±1.96±0.4
60-51	0.14± 2.13	0.32±1.73
70-61	0.58± 3.85	0.19±2.18

.) $P<0.05$(L.S.D. : 3.84

Interferon gamma (IFN-γ) also shows increased in their sera concentrations in all patients with no significantly differences ($p< 0.05$) when compared with their value in healthy persons (table 4).

Table (4) show Values of IFN-γ in patient infected with eye infection.

Age group	IFN-γ pg/ml (infected) M±SD	IFN-γ pg/ml (control) M±SD
10-2	2.13±0.13	1.58±0.14
20-11	2.26±0.43	0.1±1.43
30-21	0.01±1.86	0.21±1.49
40-31	0.05±1.69	0.12±1.40
50-41	0.09±3.42	0.18±1.42
60-51	0.08±1.87	0.12±1.66
70-61	0.41±2.67	0.1±1.76

Disscusion.

Microbes , practically bacteria form the essential causative agents of eye infection , this related with their occurrences and ability for invasion and colonization in various sites of the body (Brook et al, 2001) .

The present study revealed a total of eighty five of bacterial isolates from the three sites of eye (eye lids , conjunctiva ,dacryocystitis).

Staphylococcus aureus form the predominant bacteria of eye infection with 38.6 % of all bacterial isolates . The high percentage of S. aureus in eye infection is correlated with the normal distribution of these bacteria with healthy persons, about 20% of healthy persons harbor Staphylococci with nasal passages (Okesola ,2010). Furthermore, Staphylococci had a noticeable ability to survive in different environments and resist antibiotics and many antiseptics (Anmar ,2010) . Staphylococcus aureus contributed in eye infection and form the essential causative agents of conjunctivitis due to simplified transmissionfrom their normal habitats in upper respiratory passages to eys cross the tear ducts (Humphreys ,2012).Staphylococcus epidermidis formed the second predominant isolates of eye infection 18 isolates , most of them obtained from eye lids . However , the skin represents the normal habitat of S. epidermidis , these bacteria could isolated from several site of eye as a results of their habitat near eyes (Cervants and Mah , 2011) .

E.coli reveals a high percentage of gram negative bacterial isolates 22.7 % of all eye infection bacterial isolates . This high rate of E. coli isolates correlated with their ability to colonization and invasion to eye components in addition to the role of contamination with this agents by hands or during birth in newborns (Chen and Star ,2008 ; Quinet *et al* ,2012) .

Elisa technique pointed out only one positive case for the presence of *Chlamydia trachomatis* in eye infected infants practically in less than six months aged infant .Detection of *Chlamydia trachomatis* by cultural methods had less accuracy so, cell line and anti –Chlamydia antibody are more susceptible for detection , appearance of anti Chlamydia IgM expressed the infection of this case (Mahon and Manuselis ,2000). The low rate of this infection correlated with transmission style of this microorganism from infected mother her newborn during birth (Chojnacka *et al* , 2012) .

The present study reveals inducing of immune response due to bacterial eye infection, IL6 show elevated in their concentration in all patients . Cytokines play a critical role in imuuno regulatory processes and formation of inflammatory response , IL6 produced by monocytes and their derivites as aresults of infected of all eye infection components (Hume et al ,2006) .

Levels of IFN gamma were also increased in eye infected patients although there is no highly significantly differences ($p< 0.05$) . The secretion of interferons correlated with viral and intracellular bacteria , IFN gamma produced in early stage of infection for protection and control of disease (Horaia *et al* ,2011 ; Daio *et al* ,2012).

Conclusion.

Positive bacteria represented a clear predominant isolates from eye infection . . The level of IFN gamma in sera of eye inflammated patient show slightly enhancements , this cytokines can not be considered as a diagnostic indicator for eye inflammation**.**

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