"Cytomegalovirus Seroprevelance in Iraqi Pregnant Women"

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ABSTRACT:

BACKGROUND:

Cytomegalovirus is the commonest cause of congenital viral infection in the developed and developing countries. It is a symptomatic in 90% of infected females. Forty percent of pregnant females transmit the virus to their fetus. Ten percent of born infants whom gain the virus will get the clinical signs plus its neurological sequelae.

OBJECTIVE:

To outline the relationship between Cytomegalovirus infection among pregnant women and its influence upon their pregnancy outcome.

PATIENTS AND METHODS:

A case-control study was carried out was carried out in the teaching laboratories – medical city, Baghdad through a period from June 2010 till March 2011; upon 165 pregnant women whom taken as a patient group. Blood samples were taken from them and Cytomegalovirus-IgM antibodies plus Cytomegalovirus-IgG antibodies levels were measured via Elisa technique in both to evaluate the viral infection if present or not.

RESULTS:

The usual age whom attended the gynea-obestetric clinics were the age group from 11-19 years old 73 cases (44.24%); Next came the age group from 20-29 years old 66 cases (40.00%), thirdly was the age group from 30-39 years old 20 cases (12.12%), Lastly was the age group from 40-49 years old 6 cases (3.64%). The concentration of CMV-IgG among females whom attended clinics were seropositive in nearly half the included cases 66/165 (40.0%); while the CMV-IgM concentration was within the seronegative limits. Secondly came another group of patients with seronegative limits regarding both the CMV-IgM and CMV-IgG antibodies 56/165 cases (33.94%), next 30/165 cases (18.18%) were seropositive in their results pointing to both CMV-IgM and IgG limits; and this might be a middle point distance between the previous group and the last group were the CMV-IgM was positive and the CMV-IgG concentration was negative 13/165 cases (7.88%).

CONCLUSION:

Cytomegalovirus-IgG antibodies got important role as a protective agent against gestation abortion, if Cytomegalovirus-IgM antibodies are seropositive alone this is a risky factor to the pregnancy outcome.

KEY WORDS: cytomegalovirus, antibody, pregnancy, pregnancy outcome.

INTRODUCTION:

Human Cytomegalovirus (CMV) is a herpes virus and the most common cause of congenital viral infection and malformation in the developed countries resulting from viral intrauterine infection [1,2,3].

It is still an important perinatal virus as each year many gravidas acquired primary infection. Forty percent of cases transmitted to the fetus; and the rate of transmission highest when maternal infection has been occurred during the third trimester, but the risk of serious fetal injury is greatest when maternal infection occurs during the first trimester and early in the second trimester [4,5]. Ninety percent of infants with congenital CMV infection display no clinical manifestations at birth, the remaining 10% of intrauterine CMV infections resulting into signs at birth and they are at serious risk of long-term neurological sequelae. The risk of any sequelae in infants with symptomatic congenital CMV at birth is 90% [6,7,8,9].

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Cytomegalovirus establishes a lifelong latent infection following primary infection that can periodically reactivate with shedding of infectious virus. Primary infection, reactivation and reinfection during
pregnancy can all lead to intrauterine transmission to the developing fetus, which could lead to permanent deafness or neurological impairment [10].
There are two possible routes of transplacental transmission of CMV to the developing fetus; across syncytiotrophoblast, with subsequent infection of the underlying cytotrophoblast, or via invasive cytotrophoblast through the uterinewall[11]

PATIENTS AND METHODS:

A case-control study was carried out in the teaching laboratories – medical city, Baghdad through a period from June 2010 till March 2011; during which 165 pregnant women.In addition, a questionnaire format was performed to be asked to the included pregnant women; in order to exclude any history of other sexually transmitted disorders. All pregnant women blood specimens were taken to be investigated regarding the prevalence of anti-cytomegalovirus infection antibodies M, and G (IgM, IgG). All pregnant women who are complaining from other causes of abortion were excluded from study. After separation of blood specimens, their sera were tested to determine the concentration of CMV-IgM and CMV-IgG using Elisa technique The Elisa kits which introduced were purchased belonging to (Acon Company, U.S.A.) according to the manufacturer's instructions as follows: using a plain tubes the

blood was aspirated, labeled and allow samples to clot for 30 minutes before a centrifugation for 15minutes. Remove serum and perform the assay immediately or store samples at -20 °C for one week in order to prevent our target denaturation or degradation. Bring all kit components and samples to room temperature (22-25°C) before use, then dispense 100 µL of sample diluents' into sterile khan tubes, after that add to it 5 µL serum. Add 100 µl from the mixture to the microtiter plate wells in a duplicate manner regarding our patients and in a duplicate manner regarding the ready for use standards supplied with the kit; and the first two wells are leaved dried without any reagents addition as they regarded (blanks) wells. Cover the plate with adhesive tape supplied by the company and incubate at 37 °C for 30 minutes. Thereafter wash the microtiter wells for five times and dried carefully; followed by addition of 100 µl of conjugate to each well except the blank wells; Mix well (Mixing well in this step is important). Cover and incubate the plate for 30 minutes at 37°C. Followed by a second wash of the plate for five times also and dried carefully. After this add our substrates (A) 50 µl then (B) another 50 µl and incubate the plate at 37 °C for 15 minutes. Finally add 50 µl of the stopping solution to each well and read within 10-20 minutes the optical density (O.D.) at 450 nm using a microtiter plate reader. We calculate the cutoff value from a specific equation in order to classify our patients as comparing their values with the cutoff value of the used Elisa kit. A probability provides a quantitative description of the likely occurrence of a particular event. Probability is conventionally expressed on a scale from 0 to 1; a rare event has a probability close to 0, a very common event has a probability close to 1.

The probability of specific events (Es) calculated according to the following equation:

$$\text{Probability of event (PE)} = \frac{\text{Number of outcomes corresponding to event E}}{\text{Total number of outcomes}}$$

RESULTS:

It is clarified from Table-1 that the usual age whom attended the gynea-obestetric clinics were the age group from 11-19 years old 73 cases (44.24%) plus age group from 20-29 years old 66 cases (40.00%) with mean (average) = 41.25, median value =40, Standard deviation= 41.02.

Table1: Age distribution in Iraqi pregnant women

Females age range groups / years	Numbers	Percentage
11-19	73	44.24
20-29	66	40.00
30-39	20	12.12
40-49	6	3.64
Total	165	100.0%

Regarding Table-2; It was markedly clear from the current work that the concentration of CMV-IgG among the Iraqi females whom attended the gynea-obestetric clinics were seropositive in nearly half the included cases 66/165 (40.0%); while the CMV-IgM concentration was within the seronegative limits. Secondly came another group of patients with seronegative limits regarding both the CMV-IgM and CMV-IgG antibodies 56/165 cases (33.94%), next 30/165 cases (18.18%) were seropositive in their results pointing to both CMV-IgM and IgG limits; and this might be a middle point distance between the previous group and the last group were the CMV-IgM was positive and the CMV-IgG concentration was negative 13/165 cases (7.88%).

Table 2: Cytomegalovirus antibodies (IgM, IgG) in Iraqi pregnant women.

IgM concentration	IgG concentration	Numbers	Percentage	Events Probability Value *(P-Value)*
+	-	13	7.88%	0.08
+	+	30	18.18%	0.18
-	-	56	33.94%	0.34
-	+	66	40.0%	0.40
Total		165	100.0%	1.00

Including Table-3 in the present study; it may indicate an association between the CMV- IgG seropositivity and the Iraqi female pregnancy outcome. As 49 cases/165 their outcome were full term delivery; on the other hand only 17 cases/165 ended unfortunately with abortion before the gestation maturation. On the other hand 47/165 delivered full term babies and only 9 ended with abortion when the CMV-IgM plus IgG were seronegative; as they might be not exposed previously to the virus, in addition 24 females delivered full term babies against 6 females aborted their fetus immaturely when both CMV-IgM and IgG were seropositive; Lastly 3 females only gained their babies with full term while on the other spectrum 10 females lost their fetus improperly when the CMV-IgM was seropositive and CMV-IgG was seronegative.

Table 3: Cytomegalovirus antibodies (IgM, IgG) in Iraqi pregnant women and their pregnancy outcome

IgM concentration	IgG concentration	Full Term Delivery	*P-Value*	Gestation ended with abortion outcome	*P-Value*
+	-	3	0.02	10	0.24
+	+	24	0.2	6	0.14
-	-	47	0.38	9	0.21
-	+	49	0.4	17	0.41
Part from total sample size	123 From total 165	42 From total 165			

DISCUSSION:

Cytomegalovirus establishes a lifelong latent infection following primary infection that can periodically reactivate by shedding of infectious virus [10]

In the ongoing study it is obvious from data which obtained among group of women where CMV-IgM beside CMV-IgG are seronegative; that the risk of virus infection sequelae is negligible, but the risk of gaining primary viral infection is great; and this is run with study result was carried out by Enders *et al.* 2001 [12]

Among women group where CMV-IgM is seropositive and the CMV-IgG is seronegative; they were regarded as actively infected patients with the virus and they were mostly asymptomatic personnel; so they considered a risky targets to the community regarding the viral transmission. This agrees with a previous work done by Wreghitt *et al* 2003 [13]

Through another gained women group in the present work where the CMV-IgM was seronegative and the CMV-IgG was seropositive; they considered immunized against the virus primary infection as it already occurred previously prior to the female gestation. This is run with the same goals of a study done by Fowler 2006 [8]. So as a conclusion they decided that this situation of the mother immune response act as a protection to her fetus health. This come with agree with a study carried out by Fowler 2003 [14]

The most seriously group of this study involved women is that in which the CMV-IgM was seropositive beside the CMV-IgG was seropositive; although this means that the females are gaining acute viral infection which might be occurred in the ongoing pregnancy or it might be a reactivation to some focus of previous chronic infection.

There is only a cross reactivity of CMV-IgM to Epstein- Barr virus (EBV), measles, Herpes simplex virus, Varicella-zoster virus, and influenza vaccine; and not to CMV-IgG; which is a major issue in the development of reliable assays is the identification of viral proteins that react with antibodies induced by other viruses. Since these viruses and CMV, show considerable DNA and protein sequence homologies in conserved gene blocks, particular proteins or protein fragments have been excluded from serological a

CONCLUSION:

Cytomegalovirus-IgG antibodies got important role as a protective agent against gestation abortion, if Cytomegalovirus-IgM antibodies are seropositive alone this is a risky factor to the pregnancy outcome. ssays. Pursuing this strategy has allowed the development of recombinant tests for these viruses [14]

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