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### RESEARCH ARTICLE

## PREVALENCE OF CYTOMEGALOVIRUS IGG ANTIBODIES, POTENTIAL RISK FACTORS AND AWARENESS OF CONGENITAL CYTOMEGALOVIRUS AMONG FEMALE DOCTORS

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### ABSTRACT

Female staff in children-Mother care hospitals may run an increased risk of cytomegalovirus (CMV) contact infection leading to a congenital CMV fetopathy during pregnancy. Also, because of limited treatment options for congenital cytomegalovirus infection, preventive strategies are important so knowledge and awareness among doctors are essential for the success of preventive strategies. Thus this study was carried out to determine the positive rate of IgG among female doctors at age bearing who care for children and mothers in Sana'a hospitals. Also, to investigate the knowledge of congenital CMV among doctors involved in the study. This cross sectional study was carried out for one year (June 2017- June 2018); 178 Yemeni female doctors were included in this study. 23 (12.9%) of the total female doctors were pediatrics, 32(18%) gynecology and obstetrics, 9 (5.1%) Oto-rhinolaryngology and 114 (64%) were general practices. Blood samples were collected from all participants and tested for IgG antibodies of HCMV by ELISA. The individual's data were collected in a pre-designed questionnaire including; demographic data, risk factors. Also, a questionnaire on CMV infection was answered by doctors on the knowledge concerning epidemiology, transmission, symptoms and signs of CMV infection in adults and children; and treatment options. 13.4% of female doctors were negative (HCMV IgG <9 SU), while the rest (86.6%) were positive (HCMV IgG ≥ 11 SU). There was escalate trend of positive HCMV IgG antibodies rate with increasing age, significance association between sero-positive HCMV IgG antibodies and parity (OR=3.1, p=0.01), 1-3 pregnancy rate (OR=2.2, p=0.05), and history of surgery (OR=3.8, p=0.01). Most of the doctors were aware that most healthy adults and pregnant women do not experience any symptoms of a CMV infection (66.8%); and only one-fifth of the respondents were aware that kissing, and changing diapers, are risk factors for the transmission of CMV. Also, only half of the total respondents were aware that microcephaly (59%), and hearing loss (56.7%) could be symptoms of congenital CMV. This study revealed that the HCMV is highly endemic in our population where the prevalence of IgG among the medical female doctors was 86.5%, thus HCMV should be considered as a possible cause of morbidity and mortality in fetus and might in mothers in Yemen. Increasing knowledge and awareness is expected to enhance the prevention of transmission, to improve recognition, and to stimulate diagnostic investigations and follow-up programs.

**Keywords:** CMV, staff in children-Mother care hospitals, risk factors, knowledge, awareness.

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### INTRODUCTION

Human cytomegalovirus is a member of the *Herpesviridae* family, and the virus may be shed intermittently in bodily fluids (saliva, urine, semen, blood and breast milk). As such, its transmission

occurs both horizontally and vertically through close contact and directly from mother to embryo, fetus, or baby. Upon primary infection, which is usually asymptomatic, HCMV establishes a state of lifelong latency, during which infectious virus is difficult to isolate. Active HCMV infection can result from

primary infection in a previously sero-negative individual or reactivation in a sero-positive individual<sup>1,2</sup> in response to immune-suppression and inflammation. Viral reactivation is associated with significant morbidity and mortality in immune-compromised individuals, such as patients with HIV infection or those undergoing solid organ or bone marrow transplantation, and up to 15% of babies who acquire congenital infection, manifest signs of cytomegalic inclusion disease (CID) at birth<sup>3,4</sup>. Seroprevalence of HCMV varies from 30 to 90 % in most developed countries and the sero-prevalence is dependent on sociodemographic factors. Adult populations in Africa, Asia and South America<sup>5</sup> have higher HCMV seroprevalence than European<sup>6,7</sup> and North American populations. Vaccines are being developed for CMV<sup>8</sup>. To inform potential vaccination programmes, it is essential to understand the current epidemiology of this infection in childhood. Maternal sero-prevalence has a significant impact on the pediatric epidemiology of these infections, while children frequently transmit herpes viruses to their mothers<sup>9,10</sup>. Congenital CMV infection is the most common congenital infection worldwide, and in the developed world it is the leading non-genetic cause of sensorineural hearing loss (SNHL) in children and an important cause of neuro-developmental delay<sup>2</sup>. While awaiting treatment options, the burden of disease can be decreased by preventive strategies that reduce the risk of transmission of CMV to the pregnant woman<sup>11</sup>. A recent review of the implementation of educational hygiene interventions provides preliminary support for the positive effect of preventive strategies<sup>12</sup>. The success of preventive strategies depends on the active involvement of the doctors involved in mother and child care. Awareness of the epidemiology, transmission, diagnosis and prevention of congenital CMV is essential for every doctor. Studies report on the knowledge of women of childbearing age, and obstetricians, concerning congenital CMV<sup>13</sup>. At the current time, there is minimal information regarding the epidemiological determinants of HCMV infection in Yemen. We undertook a study to determine the prevalence of CMV antibodies level as marker for immunological status for CMV in female doctors (risk group for congenital cytomegalovirus). Also to determine general potential risk factors of contracting CMV and occupational risk factors among this group. In addition to investigate the knowledge of transmission routes of CMV, clinical manifestation and congenital sequel of CMV among doctors involved in mother and child care in Sana'a city.

## SUBJECTS AND METHODS

In the present study, a total number of 178 female doctors of hospitals in Sana'a city were enrolled. 63 (35.4%) of the total doctors were working in Al-Sabian University hospital, 16 (9%) were working in The Mother hospital, 12(6.7%) working in SMSH (YFCA), 58 (32.6%) working in Al-Thorah University Hospital and 29 (16.3%) working in Al-Kuwait University Hospital. The individual's data were collected in a pre-designed questionnaire including; demographic data,

risk factors. Also, a questionnaire on CMV infection was answered by doctors on the knowledge concerning epidemiology, transmission, symptoms and signs of CMV infection in adults and children, and treatment options were evaluated. Serum samples of female doctors were tested for HCMV IgG antibodies using standard, validated and commercially available enzyme-linked immunosorbent assay (ELISA) (Abcam's).

## Statistical analysis

To relate possible risk factors for CMV infection, the data were examined in a case-control study format. For HCMV, persons with evidence of previous or current infection with HCMV (HCMV IgG antibodies-positive) were matched up with those who were HCMV IgG antibodies negative. Differences in categorical variables were assessed using Fisher's exact tests where appropriate. Ninety-five percent confidence intervals for odds ratios were calculated according to the method of Cornfield and 95% confidence limits for simple proportions were calculated by an exact binomial method using EPI-INFO.

## RESULTS

In the present study, a total number of 178 female doctors of hospitals in Sana'a city were enrolled. The detailed results of this study are presented in Table 1, 2,3,4,5 and 6. 13.5% of female doctors were negative (HCMV IgG <9 SU), while the rest (86.5%) were positive (HCMV IgG ≥ 11 SU), 51.7% of them had high amount of HCMV IgG. There was escalate trend of positive HCMV IgG antibodies rate with increasing age, significance association between sero-positive HCMV IgG antibodies and parity (OR=3.1, p=0.01), 1-3 pregnancy rate (OR=2.2, p=0.05), and history of surgery (OR=3.8, p=0.01). Table 5 and 6 shows the number and percentage of stated Yes responses per multiple choice items on the CMV Questionnaire for all female doctors participate in the study to test knowledge concerning of HCMV clinical signs in adults, route of transmission and postnatal symptoms of HCMV. Most of the doctors were aware that most healthy adults and pregnant women do not experience any symptoms of a CMV infection (66.9%); and only one-fifth of the respondents were aware that kissing, and changing diapers, are risk factors for the transmission of CMV. Also, only half of the total respondents were aware that microcephaly (59%), and hearing loss (56.7%) could be symptoms of congenital CMV.

## DISCUSSION

Staff in hospitals may run an increased risk of cytomegalovirus (CMV) contact infection leading to a congenital CMV fetopathy during pregnancy. The main risk factor is close contact with unapparent carriers of CMV among infants etc. We therefore examined CMV sero-prevalence and possible risk factors for CMV infection among staff at a Mother-children's hospitals. To our knowledge, this is the first documented data in Sana'a city regarding the epidemiology of and knowledge of HCMV among Yemeni female doctors. According to the current study

high percentage of Anti-HCMV IgG antibodies (86.5%) among participating doctors indicated either past infection (apparently sub-clinical), current active sub-clinical disease or exposure to virus without active disease. After HCMV exposure or infection, IgG remains for the rest of life as protective antibody against the next infection. However, the primary HCMV infection needs to be elucidated either as recurrent or new infection by specific HCMV IgG avidity test.

This sero-prevalence of HCMV IgG antibody in our study (86.5%) was slightly higher than what reported in China (81.7%)<sup>14</sup> and much higher than reported in developed countries as France (51.1%)<sup>15</sup> and UK (51.5%)<sup>16</sup> among female doctors but lower to countries like Thailand, where figures of 100% sero-prevalence reported<sup>17</sup>. Also, when we compared our finding with pregnant women in previous Yemeni and Arab studies, our finding was lower than Sana'a city (100%)<sup>18</sup>; Hodeida city (98.7%)<sup>19</sup>, Taiz city (99%)<sup>20</sup>, Iraq (100%)<sup>21</sup>, Egypt (100%)<sup>22</sup>, and Sudan (97.5%)<sup>23</sup>, and also lower than that of Iran (98.8%)<sup>24</sup>, and Nigeria (94.8%)<sup>25</sup>. These high rates may be due to the poor hygienic practices and low socioeconomic status that might play significant roles in increasing the rate of HCMV exposure and infection.

Despite the general very high sero-prevalence of HCMV infection, 13.5% of female's population remained seronegative. It is likely that good hygiene, hand washing and limited sharing of edibles and used utensils serve to limit infection rates in some populations<sup>26</sup>. Also the low HCMV-IgG negative profile (13.5%) indicates that the great majority of infections occur during childhood although our data also reveal a significant increase in sero-prevalence of the female doctor's age group  $\geq 35$  years.

In our study 51.7% of female doctors showed high titer production of HCMV IgG antibodies. IgG high titer production is likely to indicate a response to endogenous viral reactivation and suggests that the more frequent exposure to viral infection within the hospitals may boost protective endogenous immunity. In this study, the age was essentially connected with CMV infection, there was escalate trend of positive HCMV IgG antibodies rate with increasing age in which the highest rate of positive HCMV IgG antibodies was in the oldest age group  $\geq 35$  years in which the positive rate was 96.2%, with associated OR equal to 4.6., and this result could be explained to debilitating of the immune system with increment in age and longer time of exposure to the virus on older people. The association between increasing age and HCMV IgG sero-prevalence positive in our study is similar to that reported elsewhere, were sero-positivity of HCMV increases steadily throughout adulthood<sup>27</sup>.

The highest rate of positive HCMV IgG antibodies was in gynecology and obstetrics doctors in whom the rate was 93.8% with associated OR equal to 2.7, followed by the rate in pediatrics in which the rate was 91.3% with associated OR equal to 1.7. In otorhinolaryngology doctors the rate of positive HCMV IgG antibodies was 88.9%. In general practice doctors the positive HCMV IgG antibodies rate was 83.3% (the

lowest) (Table 3), but the variations between these rates were statistically non-significant related to the risk of HCMV positivity, since they did not reach significant levels. This result is different from previous reports in which pediatrics were the most risk doctors because close contact with children below the age of three is considered to be the most important risk factor. Children can be unapparent CMV carriers. Over several months or years, they secrete large quantities of CMV in urine and saliva after having themselves been infected prenatally via the placenta or postnatal via breast milk or contact with other carriers, usually children. That is why, in line with current government recommendations, a ban on employment in paediatric medicine, depending on the specific area of work and activities, has to be examined particularly for female doctors at age bearing time<sup>28</sup>.

This study showed a significance association between sero-positive HCMV IgG antibodies and parity (motherhood) in which the sero-positive HCMV IgG antibodies rate was 92.2%, with associated OR equal to 3.1, CI=1.3-7.7, with significant  $\chi^2=6.5$  and  $p=0.01$  (Table 4). Parity was observed before as risk factors for increased susceptibility to acquiring CMV infection, perhaps through the direct contact with contagious secretions from their own children and poor hygiene practiced by these women<sup>29,30</sup>, so an effective prevention approach should take into account in all cases particularity in high parity families. The direct contact with infectious secretions from children of the pregnant women in addition to poor hygiene practiced by those women may increase the HCMV exposure leading to infection at any stage of pregnancy<sup>29,30</sup>, but the above situation was in disagreement to other published studies<sup>31,32</sup>.

There was significance association between sero-positive HCMV IgG antibodies and history of surgery in which the sero-positive HCMV IgG antibodies rate was 94.4%, with associated OR equal to 3.8, CI=1.3-11.7, with significant  $\chi^2=6.2$  and  $p=0.01$ . Most surgery accompanied with blood transfusion infection which may lead to significant obstacles in immune-compromised persons. This is similar to study that carried out in Kenya<sup>21</sup>. However, in dissimilarity to other studies in Mexico and Nigeria<sup>33,34</sup>.

In the this study the following variants like history of miscarriage, infection, blood transfusion and cupping were not related to the risk of HCMV positivity, since they did not reach significant levels. However, some authors reported a significant association between history of miscarriage and blood transfusion with HCMV IgG seropositivity<sup>35,36</sup>.

We investigated the knowledge of congenital CMV infection among female doctors in Sana'a city involved in mother and child care. Several interesting findings were shown. First, doctors seemed to miscalculate the chance of encountering a child with congenital CMV infection in medical practice. There seemed to be a risk of under diagnosis, since the prevalence of congenital CMV, internationally do not estimated sometimes overestimated, but more frequently underestimated by respondents in this study (only 21.3% answer yes for possible occurring congenital HCMV in Yemen).

Secondly, preventive strategies are assumed to be effective only when doctors are sufficiently well informed to advise their patients properly. The data in this study show that most of the doctors were aware that most healthy adults and pregnant women do not experience any symptoms of a CMV infection (66.8%). Worryingly, however, only one-fifth of the respondents, including those working with pregnant patients, were aware that kissing, and changing diapers, are risk factors for the transmission of CMV.

Thirdly, including congenital CMV infection in the differential diagnosis in symptomatic newborns is crucially important. Since only half of the total respondents in this study were aware that microcephaly (59%) and hearing loss (56.7%) could be symptoms of congenital CMV, it seems possible that these children may be left undiagnosed, with possibilities for treatment and follow-up not explored. It is worrying that only 20.2% of all respondents realized that congenital CMV frequently does not give rise to any symptoms and signs at birth, and that 14% of these asymptomatic newborns will develop long-term sequelae.<sup>37</sup> Finally, we were surprised that 45% of the respondents thought that the antiviral therapy of newborns with congenital CMV infections is common practice in the world. Even though antiviral therapy has been shown to prevent hearing deterioration in newborns with symptomatic CMV infections, this practice is not yet widespread worldwide<sup>38</sup>.

In the case of CMV it is especially important that doctors involved in the care of women who are or who may become pregnant are able to advice on the risk of congenital CMV and how this risk may be reduced. To date, information on congenital CMV is not regularly included in preconception and antenatal consultations.

## CONCLUSION

This study revealed that the HCMV is highly endemic in our population where the prevalence of IgG among the medical female doctors in age bearing was 86.5%, thus HCMV should be considered as a possible cause of morbidity and mortality in fetus and might in mothers in Yemen. Increasing knowledge and awareness is expected to enhance the prevention of transmission, to improve recognition, and to stimulate diagnostic investigations and follow-up programs.

## RECOMMENDATION

Control measures should be applied in hospitals as well as routine investigation for HCMV in pregnant women. Also vaccine, prophylactic and pre-emptive strategies should be developed.

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## AUTHOR'S CONTRIBUTION

This research work is part of A M.Sc. thesis. The candidate is the first author (MAA) who conducted field works, laboratory works and wrote up the thesis. The corresponding author (HAA) supervised the experimental work, revised and edited the thesis draft

and the manuscript. (AGM) was co-advisor of the work, and (SMND) helped the hospital works.

## CONFLICT OF INTEREST

"No conflict of interest associated with this work".

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**Table 1: The levels of HCMV IgG antibodies among female doctors in Sana'a city, Yemen, 2018**

Level titers (Standard Units)	The levels of HCMV IgG antibodies	
	Number	%
Negative (<9 SU)	24	13.5
Low positive (11-26 SU)	11	6.2
Medium positive (27-42 SU)	51	28.6
High positive (> 42 SU)	92	51.7
Total	178	100

SU=Standard Units, Cut-off: 10 SU, Grey zone: 9-11 SU= repeated, Negative: <9 SU, Positive: >11 SU

**Table 2: The seroprevalence of HCMV IgG antibodies in relation with age of female's participants**

Age groups	HCMV IgG positive (n = 154)		OR	CI	$\chi^2$	P
	No.	%				
≤30 years n=29	21	72.4	0.31	0.1-0.8	5.9	0.01
31-34 years n=123	108	87.8	1.4	0.6-3.4	0.56	0.45
≥35 years n=26	25	96.2	4.6	0.8-34	2.4	0.11
Total n=178	154	86.5				

OR-Odds ratio = Relative risk, CI-Confidence intervals,  $\chi^2$ -Chi-square = 3.9 or more significant, PV-Probability value = 0.05 or less is significant

**Table 3: The seroprevalence of HCMV IgG antibodies in relation with specialty of work for female doctors**

Field of works	HCMV IgG positive (n = 154)		OR	CI	$\chi^2$	P
	No.	%				
Pediatrics n=23	21	91.3	1.7	0.3-7.9	0.5	0.47
Gynecology and obstetrics n=32	30	93.8	2.7	0.6-11.9	1.8	0.18
Oto-rhinolaryngology n=9	8	88.9	1.2	0.1-10	0.045	0.8
General practice n=114	95	83.3	0.4	0.1-1.1	2.8	0.09
Total n=178	154	86.5				

OR-Odds ratio = Relative risk, CI-Confidence intervals,  $\chi^2$ -Chi-square = 3.9 or more significant, *p*-Probability value = 0.05 or less significant

**Table 4: The association between sero-positive HCMV IgG antibodies with host factors of female doctor participants.**

Factors	HCMV IgG positive (n = 154)		OR	CI	$\chi^2$	P
	No.	%				
Motherhood (parity)						
Yes n= 102	94	92.2	3.1	1.3-7.7	6.5	0.01
No n= 76	60	78.9	0.3	0.12-0.7	6.5	0.01
Number of pregnancy ended with live child						
1-3 n= 89	81	91	2.2	1-5.5	3.9	0.05
<3 n=13	13	100	Undefined		2.1	0.13
Spontaneous Abortion history of miscarriage						
n=19						
1-3 n= 16	14	87.5	1.1	0.23-5.1	0.041	0.9
<3 n= 3	2	66.6	0.3	0.02-3.5	1.03	0.3
History of infection diseases n= 35						
Blood transfusion n=19	17	89.5	1.3	0.3-6.3	0.15	0.67
Cupping n= 4	3	75	0.45	0.04-4.5	0.46	0.49
Surgery n= 71	67	94.4	3.8	1.3-11.7	6.2	0.01

OR-Odds ratio = Relative risk, CI-Confidence intervals,  $\chi^2$ -Chi-square = 3.9 or more significant, *p*-Probability value = 0.05 or less significant

**Table 5: The number and percentage of stated Yes responses per multiple choice items on the CMV Questionnaire for all female doctors participate in the study to test knowledge concerning of HCMV**

Knowledge concerning	Number	Percentage
Is HCMV problem in Yemen (Yes)	37	21
Transmission route		
True answers	78	43.8
Kissing		
Changing diapers	39	21.9
Breast milk	53	29.8
Blood contact	82	46.1
Sexual intercourse	38	21.3
False answers	101	56.7
Air conduction		
Direct skin contact	39	21.9
Symptoms in immune competent adults		
True answers		
No symptoms	119	66.8
Not feeling well	59	33.2
Fever	59	33.2
Elevated liver enzymes	37	20.6
False answers	9	5.1
Cardiac problems		
Thrombosis	7	3.9
Visual problems	22	12.4

**Table 6: The number and percentage of stated Yes responses per multiple choice items on the CMV Questionnaire Postnatal symptoms for female doctors in Sana'a city hospitals**

Knowledge concerning	Number	Percentage
Postnatal symptoms		
True answers		
No symptoms	36	20.2
Petechiae	91	51.1
Microcephaly	105	59
Seizures	28	15.7
False answers		
Heart defect	59	33.1
Macrosomia	5	2.8
Renal problems	28	15.7
Anal atresia	3	1.7
Long-term effects		
True answers		
Hearing loss	101	56.7
Cognitive delay	85	47.8
Motor delay	51	28.6
Seizures	23	12.9
Autism	2	1.1
Visual problems	58	32.6
False answers		
Obesity	3	1.7
Increased risk for malignancy	26	14.6
viable and use of Antiviral therapy worldwide (Yes)	80	45