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Pattern of Cytomegalovirus among Yemeni Population

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Abstract

The Cytomegalovirus is highly prevalent worldwide, the prevalence varied according to country and the epidemiological factors and ranged from 100% in both Africa and Asia to 80% in Europe and North America. CMV establishes lifelong latency that can lead to periodic reactivation spontaneously or in conditions where immunity is suppressed. In addition, CMV can be easily transmitted from person to person by direct contact, with saliva, semen, vaginal fluids, blood, urine, tears, feces, and maternally from pregnant women to infant or by breast milk feeding. CMV infections are could be a symptomatic or asymptomatic. In fetuses CMV can cause severe disease and disabilities with mortality rate 20% to 30% and 10% to 15% of congenitally infected infants will have symptoms at birth. In Yemen the prevalence rate of CMV IgG among pregnant women ranged from 94.6% to 98.7%. Compared from 97.9% to 100% among healthy adult male and female were seropositive for CMV IgG respectively. In our country methods of prevention and protection against CMV are still completely absent, as blood is transfused without screening blood donors for CMV, which has exacerbated of the problem and required us to conduct in-depth and more comprehensive studies to highlight the problem regarding CMV. So we aimed this study to determine the pattern of Cytomegalovirus among Yemeni population.

Material and Method This is a cross-sectional study was conducted separately on four dotages from 2021 to 2024 in Taiz governorate at AL-Turbah City and in Sana'a Capital City-Yemen. Blood specimens were collected from participants. The serum specimens were subjected for detection anti=CMV (IgG and IgM) by Electro-Chemiluminescence Immunoassay according to manufacturer's instructions.

Result A total 920 enrolled cases, 847 (92.1%) were seropositive for either of IgG or IgM or for both of (IgG and IgM) antibodies. Of these 835 (90.8%) and 141 (15.3%) were seropositive for anti-CMV IgG and IgM antibodies respectively. Approximately similar prevalence rate of anti-CMV Ab in Sana'a capital city (92.6%, OR = 1.2 and P = 0.542), Taiz governorate (91.5% OR = 0.9 and P = 0.542)), urban (92.6% OR = 1.2 and P = 0.513) and rural (91.4%, OR = 0.9 and P = 0.513) areas, where, the differ

ence in the prevalence of anti-CMV Ab was statistically insignificance but the risk of infection was found higher among urban. Significantly high prevalence of anti-CMV antibodies with increasing in the risk of infection was found among married (93.0%, OR=3.4 and P=0.000), individuals who had blood transfusion (99.2%, OR=9.2 and P=0.008), individual who had dental procedures (99.5%, OR=23.0 and P=0.000), illiterate (98.9%, OR=8.6 and P=0.010), and manual workers (95.8 %, OR=2.2 and P=0.033).

Conclusions: Cytomegalovirus is a silent and neglected health problem and will still ongoing as one of the most important health problems among population in our country.

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Introduction

Cytomegalovirus (CMV) is a widespread virus belongs to the β-herpes virus which can infect a large proportion of the human population worldwide. Like all herpes viruses CMV establishes lifelong latency that can lead to periodic reactivation spontaneously or in conditions where immunity is suppressed. People can also be infected with different strain of CMV in their lives [1]. Anti-Cytomegalovirus IgM antibodies (anti-CMV IgM Ab) produce after primary infection directly, while anti-CMV IgG antibodies produce after occurrence of infection. CMV IgM Ab remains for three or four months, while anti-CMV IgG Ab remains lifelong [2]. Infection by cytomegalovirus does not have a cure and there is no vaccine available for it [3].

Cytomegalovirus is highly prevalent worldwide with prevalence varied according to country and the epidemiological factors with an account from 80% in Europe and North America to 100% in both Africa and Asia [4]. However, Zuhair reported that the highest seroprevalence was seen in the World Health Organization (WHO) Eastern Mediterranean region 90%: (85-94) and the lowest in WHO European region 66% (56-74), while, Heredia found a 1.4-fold increase in CMV seroprevalence in the last 10 years [1,5,6].

The seroprevalence of CMV among women in reproductive age with rates ranging from 40 % in developed countries to over 90% in developing countries [7-9].

The virus can spread from person to person through

contact with saliva, semen, vaginal fluids, blood, urine, tears, feces, and breast milk. Most CMV infections are a symptomatic in healthy individuals. Although the infection is often asymptomatic or vague, it may present with flu-like symptoms including fever, myalgia, lymphadenopathy and fatigue. After causing primary infection, viral components can be found in many body fluids such as urine, saliva, vaginal secretions and breast milk for months. In fetuses CMV can cause severe disease and disabilities such as sensorineural deafness, optic atrophy, myocarditis and mental retardation [10]. On other hand, it was reported that, 10% to 15% of congenitally infected infants will have symptoms at birth including intrauterine growth restriction, microcephaly, hepatosplenomegaly, petechiae, jaundice, retinitis, intracerebral calcifications, thrombocytopenia, and anemia, and 20% to 30% of them will die, mostly of disseminated intravascular coagulation, hepatic dysfunction, or bacterial super infection [11]. Most of the congenitally infected infants (85-90%) have no signs or symptoms at birth, but 5% to 15% of them will develop sequelae such as sensorineural hearing loss, delay of psychomotor development, and visual impairment, Ten percent of infected infants are symptomatic at birth; approximately one third of them will die and up to 90% of the survivors will develop long-term sequelae [12,13,14].

The fetus can be infected by either a newly acquired maternal infection or recurrent (reactivation) mater

-nal infection [3]. Maternal acquisition of infection might resemble from multiple ways including sexual and non-sexual contact, blood products and organ transplant [15].

It was found that, the risk of intrauterine transmission is highest when primary infection occurs during pregnancy, with a higher rate of vertical transmission in mothers with older gestational age at infection, while the risk of adverse fetal effects significantly increases if fetal infection occurs during the first half of pregnancy [16-18]. Breastfeeding may represent risk factors associated with increased CMV transmission to the child [19].

In Yemen, our problem is with diseases in general, and infectious and transmissible diseases in particular, and those that result in health complications for diseases and increase the rate of morbidity and mortality in general, and in particular, bacterial and viral microorganisms transmitted from mothers to fetuses and newborns, causing high mortality, miscarriages, and permanent disabilities for children, affecting their lives and the lives of their families for decades. and having negative social and economic effects on the individual, society, and the state. As a result of the lack of research work in some medical fields and its scarcity, and even its absence in most medical specialties, we find ourselves as researchers facing closed doors that need to be knocked on and then opened to look at what is behind those doors. In fact, in most cases, we are unable to determine which door to start from and where our priority to start, because in reality everything is a priority due to the lack of studies and researches to start on the basis of them. This is what makes me facing many questions that confront me with every research I conduct it, and we even find ourselves as if we were at zero position and that we need to determine our direction in which we should proceed. An example of this is that if I depend on a specific number of people, a specific place, and a specific category of society to conduct my research, I will find myself with open questions and the results of my research will raise more questions than they answer, therefore we aimed to conduct this study.

In Yemen, our problem is diseases in general, and infectious and contagious diseases in particular, and those that can lead to health complications and

increase morbidity and mortality rates, including microbes that are transmitted from mothers to fetuses and newborns, causing high rates of death, miscarriage and disabilities for children, affecting their lives and the lives of their families for decades, and having negative social and economic effects on the individual, society and the state. As a result of the scarcity of research work in some medical fields and its rarity, and even its absence in most medical specialties, we find ourselves as researchers facing closed doors that need to be knocked on and opening them to look beyond them. In fact, most of the time, we cannot determine which door to start from and where to start, because everything is a priority due to lack of studies and research guiding us on which one to start. This is what makes me face many questions that I face with every research I do, and we even find ourselves as if we are at point zero, and that we need to determine the direction in which we should go. An example of this is that if I depend on a specific number of people, a specific place, and a specific category of society to conduct my research, I will find myself facing open questions, and the results of my research will raise more questions than they answer, and that is why we aimed to conduct this study.

In Yemen, a few studies have investigated CMV infection in different classes of population and geographic areas. These studies revealed that the prevalence rate of CMV IgG among pregnant women ranged from 94.6% to 98.7 %. Compared to100% and 97.9% of adult health male and female were seropositive for CMV IgG respectively [20-23]. While 96.6 % of blood donors were seropositive for IgG and 5.5 % were seropositive for CMV IgM [24]. Low prevalence (86.6%) of CMV IgG reported among women doctors [25]. Preventive measures to reduce the incidence of CMV-infections have to be based on assessment of risk factors for CMV-infection [26].

In our country, methods of prevention and protection against herpes viruses in general and cytomegalovirus in particular are still completely absent, as blood is transfused without screening blood donors for CMV, which has exacerbated of the problem. In addition, in previous decades there is lack of studies, as the efforts of health institutions and researchers have focused on studying hepatitis and immunodeficiency viruses and a neglect problem regarding cytomegalovirus.

In the last decade, few studies have been conducted which have given us an indication of the high prevalence of cytomegalovirus which became a silent and neglected virus in Yemen. On other hand, it is crucial to consistently monitor and gather data on cytomegalovirus seroprevalence from different regions to be considered for future public health strategies and interventions. Where we intend to make a modest contribution through this study to highlight on the problem and knowledge on CMV distribution in order to draw attention of authorities and those interested to start taking precaution and preventive measures to reduce the disease and help future seeking for vaccination against virus. Therefore, we aimed to conduct this study, which was carried out on stages over four years and in more than one region and targeting different groups of people and to study the effect of demographic characteristics on prevalence of CMV in Taiz and Sana'a Capital city in Yemen.

Material and Method

This is cross-sectional study was conducted separately on four dotages from 2021 to 2024 in Taiz governorate at AL-Turbah city from January to March 2021 and from October 2021 to January 2022 and in Sana'a Capital city from March to august 2022 and from October 2023 to January 2024 as graduation research project by researcher and students of medical laboratory departments Faculty of Medical and Health Sciences Taiz university AL-Turbah branch, Faculty of Medical and Health Sciences Yemeni Jordanian University and, Faculty of Medical and Health Sciences Civilization University. Fall information of each studied individuals of the targeted groups was taken and recorded in a predesign questioner, including socio demographical characteristics. The purpose of the study was explained briefly to each selected voluntary. Verbal consent taken from each the subject had right to withdraw from study in any time without giving any reason.

Method

About 5 ml of venous blood was collected aseptically from each selected cases into sterile test tube without anti-coagulant labeled properly with a patient identification number. Serum was separated by using micropipette following centrifugation at 3000g for 5 minutes, to Eppendorf tubes, each which are stored in deep freeze refrigerators at -28°C for detection an

-anti-CMV IgG and IgM . The serum specimens of all participates were subjected for detection anti=CMV (IgG, IgM) by Electro-Chemiluminescence Immunoassay (ECLIA) technique (Cobas e411 analyzer, Roche Diagnostic GmbH, Mannheim, Germany) and by enzyme linked immunosorbent assay techniques (ELISA) using monocent kit CA 91304, monocent, Inc, USA, according to manufacturer's instructions.

Statistical Analysis

All statistical analysis was done using the statistical science (SPSS) software package version 20 (USAT).

Results

Anti-CMV Ab (IgG + IgM) Antibodies

We collected 460 cases from Taiz governorate in study conducted from January to March 2021and from October 2021 to January 2022, and 460 cases from Sana'a capital city in study conducted from March to August 2022 and from October 2023 to January 2024. A total 920 enrolled cases in this study were screened for anti-CMV antibodies. Among them 847 (92.1%) were seropositive for either or both of anti-CMV IgG and IgM antibodies. of these 835 (90.8%) and 141 (15.3%) were seropositive for anti-CMV IgG and IgM antibodies respectively.

Significantly high prevalence of anti-CMV antibodies with increasing in the risk of infection was found among married (93.0%, OR=3.4 and P=0.000), blood transfusion (99.2%, OR=9.2and P =0.008), dental procedures (99.5%, OR=23.0 and P =0.000), illiterate (98.9%, OR=8.6 and P =0.010), and manual workers (95.8 %, OR=2.2 and P=0.033). Meanwhile, higher prevalence of anti-CMV antibodies was identified among individual who had blood transfusion (99.2%), and exposed to dental procedure (99.5%), while, low prevalence rate of anti-CMV antibodies was found among single with an account 80.4%. On other hand, the risk was found higher with no significance in the difference rate of anti-CMV antibodies among females as compared to males and subject aged 26-35 and 36-45 years old as compared to others age groups. In addition the risk was found higher among primary level of education and house wife.

Approximately similar prevalence rate of anti-CMV Ab in Sana'a capital city (92.6%, OR= 1.2 and P=0.542), Taiz governorate (91.5% OR= 0.9 and P=0.542)), and

among residing urban (92.6% OR= 1.2 and P=0.513) and rural (91.4%, OR=0.9 and P=0.513) areas, where, the difference in the prevalence of anti-CMV Ab was statistically insignificance but the risk of infection was found higher among urban. Similar observation in prevalence rate of anti-CMV Ab among males (91.0% OR= 0.3 and P=0. 294) and females (92.9% OR= 1.3 and P=0. 294) with more risk among females as compared to males. it was found that, the rate of anti-CMV Ab rises with increasing in age where, 89.1%89.1, 92.7% and 95.5% of subject aged less than 25%, 26-35 and 36 -45 36 -45 were seropositive for CMV.

Anti-CMV IgG Antibodies

The result of our study revealed that, 835(90.8%) of cases had of anti-CMV IgG Ab. however, present study demonstrated approximately similar significantly high prevalence of anti-CMV IgG antibodies with increasing risk of infection among married (91.9%, OR=2.7:and P =0.000), blood transfusion (96.9 %, OR=3.5 and P =0.027), dental procedures (98.1 %, OR=6.5 and P =0.000), illiterate (96.9%, OR=3.3 and P=0.037), and manual workers (94.7%, OR=2.1and P=0.034).

Anti-CMV IgM Antibodies

Significantly high anti-CMV IgM Ab was found among females (21.7 %, OR=3.7 and P=0.000) house wife (21.4 %, OR=2.7 and P=0.000), Illiterate (31.5 %, OR=2.9 and P=0.000) primary level of education (24.5 %, OR=2.4 and P=0.000), cases who had blood transfusion (45.4%, OR=6.2 and P=0.000), and marital stratus (16.8 %, OR=9.2 and P=0.000). Low anti-CMV IgM Ab was found among non-marital status and high educated individuals with an account 2.2% and 8.4 % respectively.

Discussion

Cytomegalovirus is a virus that causes a lifelong latent infection. Therefore, positive cases for IgG and negative for IgM do not mean that they are not infected by the virus, but rather infected cases, as the virus remains latent in the body of infected persons for life.

Anti-CMV Ab (IgG + IgM)

The prevalence rate of anti-CMV Ab in this study (92.1%) was in consent with that reported in Saudi Arabia 92.1%, Taiwan 91.1% and approximate

similar 94.1% to that reported in Korea [26,27]. However, our study demonstrates low prevalence of anti-CMV Ab than that reported in Taiz City (99%) and Hodeida City (98.7%) which could be related due to all previous study conducted on pregnant women [28,19,21]. On other hand, high anti-CMV Ab reported in studies conducted in Iraq (100%), and Palestine (96%) [29,30]. While, our study yielded higher prevalence rate than that reported in study conducted in Japan 69.7 % and in Kenya 77 % [31,32].

Several studies revealed high prevalence the rate of anti-CMV Ab among females as compared to males, where, the result of study conducted by Josheghani and Wadhah et al, demonstrated high anti-CMV Ab prevalence among females with an account 97% and 100% respectively [21,32-35,23].

Flanders et al, reported less (22.0% – 61%) prevalence of anti-CMV among age groups as compared to present study, while, Josheghani et al, reported higher anti-CMV Ab rate (97.4%) where, the prevalence of CMV increase with age which was found similar to that reported by Flanders et al, and Almoaish et al, which could be explained due to that, the effective control of CMV is impaired during healthy ageing, most probably due to loss of cellular control of early viral reactivation [34,35,25].

Present study revealed high risk with significance highest (99.2% OR = 9.2 & P =0.008) anti-CMV Ab prevalence rate among cases who had blood transfusion, which could be explained that blood is transfused to patients without screening blood donors for CMV.

Anti-CMV IgG Ab

The result of recent study revealed (90.8%) anti-CMV IgG which was hound more than that reported by Alsafi and Alghamdi (88.4%) [36]. Likewise, recent study showed more CMV IgG seropositive than those studies conducted in Germany, France and in Bulgaria, with an account 42.3%, 41.9% in and 85% respectively [37-39]. While, the present study yield low prevalence than that reported by Al-musawi and Khairi et al, Jin Q et al, and Alghalibi whom showed that, 93.2%, 97.5%, 95% and 98.7%were positive for anti-CMV IgG respectively [40-42, 21].

As seen in the table 3, approximate similar prevalence

rate of anti-CMV IgG among males (90.2 %) and female (91.2%) but the risk was higher among female which was similar to that reported by Choi et al [28].

In Yemen it was reported that, 100% and 97.9% of males and females were seropositive for anti- CMV IgG antibodies respectively compared to 86.6% of Yemeni female doctors were positive for CMV IgG reported by Almoaish et al [21,25].

The rate of anti-CMV IgG increased with age was similar to that reported by Choi et al, and Al-Sabri et al, and high prevalence 100% of IgG among illiterate and primary education was reported by Mohammadi et al [24,28,43].

Jin et al, reported that, CMV IgG prevalence increased significantly with age (P < 0.01) [42].

Women living in downtown areas showed higher IgG prevalence than those residing in urban areas (p = 0.023) [44]. As compared to recent study, Khairi et al, reported high anti- CMV IgG antibodies among illiterate (100%), primary school (98.8%), intermediate school (97.7%), secondary school (97.8%) and high education (100%) compared to 43.6% of non-workers were seropositive for anti-CMV IgG compared to 84.7% in our study [41].

Anti-CMV IgM Ab

The overall Anti-CMV IgM in our study was 15.3% which was found equivalent to that reported in In Pakistan (12.71%) and more than that reported by Alghalibi et al, (1.8%) [44,21]. AbJumaan et al (3.8%), Jin et al, (1.28%) and Khairi et al, (6.0%) [41,42,22].

Our study revealed non significance and approximately similar anti-CMV IgM prevalence among those who residing in rulers (15.0%, OR=0.9:and P=0.801) and urban (15.6%, OR=1.0:and P=0.801) areas which was found in disagreed with that reported by Jin et al, who showed, that the seroprevalence of anti-CMV IgM was 1.28%. and the prevalence of anti-CMV IgM among those residing in rulers' areas showed higher IgG prevalence than those residing in urban areas (p = 0.023). On other hand, approximate similar high anti-CMV IgM 13.6% was reported by Alsafi and Agamid [42,32]. Although high (92.1%) prevalence anti-CMV IgG and IgM, but not encourging

that they have an immunity against CMV, as cytomegalovirus is a virus that can establish lifelong latency and immune individuals can experience repeat infections from endogenous (reactivation) or exogenous (reinfection) sources despite the host committing substantial immune resources against CMV [45].

Conclusions

- 1. Cytomegalovirus is a silent and neglected virus in Yemen.
- 2. High (92.1%) CMV prevalence rate will still ongoing as one of the most important health problems among population in general and among pregnant women and newborn infants in particular.

Recommendations

- 1. More comprehensive study including prevalence rate and disabilities among newborn infants and children
- 2. Screening blood donors.
- 3. Precaution and preventive measures must be to taken in considerations of health institutions and health workers.
- 4. Seeking for vaccination against virus.

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Table 1: Frequency of Anti-CMV-IgM and IgG among Participants

Result		Ig G	Total				
		Positive		Negative		Total	
		N	%	N	%	N	%
Ig M	Positive	129	91.5	12	8.5	141	15.3
	Negative	706	90.6	73	9.4	779	84.7
Total		835	90.8	85	9.2	920	100.0

Table 2: Seropositive of anti-CMV (IgG +IgM) among participants

Demographic ch	Positive N = 847 (92.1%) N %		X ²	OR: (CI)	P	
Governorates	Sana'a n=460	426	92.6	0.4	1.2: (0.6-1.4)	0.542
	Taiz n=460	421	91.5	0.4	0.9: (0.7-1.9)	0.542
Residency	Urban n=500	463	92.6	0.4	<u>1.2</u> (0.7-1.9)	0.513
	Rulers n=420	384	91.4	0.4	0.9: (0.6-1.4)	0.513
Gender	Male n= 400	364	91.0	1.1	<u>0.3</u> : (0.5-1.3)	0. 294
Gender	Female n=520	483	92.9	1.1	1.3: (0.8-2.1)	0. 294
Marital status	Yes n=827	772	93.3	18.5	3.4:(1.9-6.0)	0.000*
Mai itai status	No n=93	75	80.4	18.5	0.3:(0.2-0.6)	0.000
	≤25 n=258	230	89.1	4.1	0.6: (0.4-1.0)	0.041
A	26-35 n=480	445	92.7	0.6	1.2:(0.8-1.9)	0.451
Age	36 -45 n=155	148	95.5	3.0	2.0: (0.9-4.4)	0.084
	≥ 45 n=27	24	88.9	0.4	0,7: (0.2-2.3)	0.535
Blood	Yes n=126	126	99.2	7.0	9.2 (1.3-68.9)	0.008*
transfusion	No n=794	721	90.9	7.0	0.1: (0.01-0.6)	0.008
Dental	Yes n=206	205	99.5	20.2	23.0 (3.1-168.5)	0.000*
Dentai	No n=714	642	89.9	20.2	0.04 (0.01-0.3)	0.000
4	Illiterate n=92	91	98.9	6.6	8.6: (1.2-83.1)	0.010*
Education	Primary n=245	232	94.7	3.1	1.7: (0.9-3.2)	0.076
Education	Secondary n=321	293	91.3	0.4	0.9 (0.5-1.4)	0.517
	High n=262	231	88.2	7.6	0.5: (0.3-0.8)	0.006*
	House wife n=467	435	93.1	1.5	1.4: (0.8-2.2)	0.217
Occupation	Employee n=191	169	88.5	4.2	0.6: (0.3-1.0)	0.040
Occupation	Manual worker n = 190	182	95.8	4.5	2.2: (1.1-4.7)	0033
	Non n= 72	61	84.7	5.8	0.4: (0.2-0.9)	0.016

^{*} Statistically significant, χ 2: Chi-square, OR: Odd ratio, N: number, CI: Confidence interval, p: probability. (χ 2 \geq 3.84, p < 0.05: significant).

Table 3: Seroprevalence of Anti-CMV IgG and Anti-CMV IgM) among Participants

Demographic characteristics		Positive <u>IgM</u> : N = 141 (15.3%)				Positive IgG: N = 835(90.8%)				
		N (%)	X ²	OR: (CI)	P	N (%)	X ²	OR: (CI)	P	
Governorates	Sana'a n=460	71(15.4.)	0.01	10: (0.7-1.5)	0.927	418 (90.9)	0.01	10: (0.6-1.6)	0.909	
	Taiz n=460	70 (15.2.)	0.01	0.98 (0.7-1.5)	0.927	417(90.7)	0.01	0,97: (0.6-1.5)	0.909	
Residency	Urban n=500	79(<u>15.6.)</u>	0.1	11: (0.7-1.5)	0.801	454(<u>90.9.)</u>	0.002	<u>10:</u> (0.7-1.6)	0.964	
	Rulers n=420	63(15.0.)	0.1	0.9: (0.7-1.4)	0.801	381(90.7)	0.002	0.99 (0.6-1.6)	0.964	
Gender	Male n= 400	28(70)	37.9	03:(0.2-0.4)	0.000	361(90.2)	0.2	0.9 (0.6-1.4)	0.639	
	Female n=520	113(21.7)	37.9	3.7: (2.4-5. <u>7.)</u>	0.000*	474(912)	0.2	<u>11:</u> (0.7-1.7)	0.639	
Marital status	Yes n=827	139(16.8.)	13.8	92:(2.2-37.8)	0.000*	760(91.9.)	12.6	2.7: (1.5-4.8)	0.000	
	No n=93	2(2.2)	13.8	0,1: (0.2-0.5)	0.000	75(80.6)	12.6	04:(0.2-0.7)	0.000	
	≤ 25 n=258	41 (15.9)	0.1	1.1 (0.7-1. <u>6.)</u>	0.766	226(87.6)	4.3	<u>0.6.</u> (0.4-1.0)	0.039	
	26-35 n=480	71 (14.8.)	0.2	<u>0.9 :</u> (0.6-1.3)	0.638	438(91.2)	0.3	11: (0.7-1.8)	0.593	
Age	36 -45 n=155	25 (16.1.)	0.1	11: (0.7-1.7)	0.761	147(948)	3.7	2.0: (1.0-4.4)	0.055	
	≥ 45 n=27	4(148)	0.01	10: (0.3-2.8)	0.940	24(88.9.)	0.1	0.8:(0.2-2.7)	0.733	
Blood transfusion	Yes n=97	44(45.4)	76.4	62:(3.0-9.8)	0.000*	94(969)	4.9	3.5: (2.3-17.9)	0.027	
	No n=823	97 (11.8)	0.2	0.1 (0.1-0.3)	0.000	741(90.0)	4.9	0.3 (0.1-0.9)	0.027	
Dental	Yes n=206	38 (18.4.)	2.0	13: (0.9-2.0)	0.159	202(981)	16.9	6.5. (2.3-17.9)	0.000	
	No n=714	103(14.4)	2.0	0.8: (0.5-1.1)	0.159	633(887)	16.9	0.2 (0.1-0.4)	0.000	
Education	Illiterate n=92	29 (31.5)	20.6	29:(2.8-4.8)	0.000*	89(969)	4.4	33:(1.0=10.5)	0.037	
	Primary n=245	60 (24.5)	21.6	<u>2.4:</u> (1.8-3.4)	0.000*	228(931)	2.1	<u>15:</u> (0.9-2.6)	0.147	
	Secondary n=321	30 (9.3.)	13.7	0.5: (0.3-0.7)	0.000	288(897)	0.6	0.8: (0.5-1.3.)	0.425	
	High n=262	22 (84)	13.6	04:(0.37)	0.000	230(87.8)	3.9	0.6: (0.4-1.0)	0.049	
Occupation	House wife n=467	100 (21.4)	27.1	27:(1.9-4.0)	0.000*	428(91.6)	0.9	12: (0.8-1.9)	0.345	
	Employee n=191	22(11.5)	2.7	0,7: (0.4-1.1)	0.101	166(86.9.)	4.3	0.6:(0.4-1.0)	0.039	
	Manual worker n=190	13(6.8)	13.4	0.3:(0,1-0.6)	0.000	84(947)	4.5	2.1: (1.0-4.1)	0.034	
	Non n= 72	6(83)	2.9	0.5:(0.2-1.1)	0.086	61(847)	3.4	0.5:(0.3-1.1)	0.065	

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