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Can cytomegalovirus infection affect male reproductive function? Results of a retrospective single-centre analysis

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Abstract

Our objective was to investigate whether the chronic cytomegalovirus (CMV) infection can affect semen parameters in men with couple infertility and to assess the impact of male CMV infection on the reproductive outcomes of CMV-seronegative women suffering from tubal factor. Group 1 included CMV IgG-seropositive men, Group 2 CMV IgG-seronegative patients. Seminal parameters, two-pronuclear (2PN) fertilization rate (FR), 1-2-3PN FR, cleavage rate (CR), miscarriage rate (MR), pregnancy rate (PR) and live birth rate (LBR) were collected. Two hundred and twenty-two men were included: 115 (51.8%) in Group 1 and 107 (48.2%) in Group 2. There was reported a low trend towards higher sperm concentration/ml, total sperm count and viability in CMV IgG-seronegative males, compared to CMV IgG-seropositive ($p > .05$). Semen volume, pH, motility and normal sperm morphology were similar among groups. Considering the subgroup of men, partners of CMV IgG-seronegative females, 65 couples (29.2%) were selected. Median 2PN FR was 67%, total FR 83%, CR 100%, PR/cycle 26.2%, MR 10.8%, LBR/cycle 15.4%. No significant differences were found regarding the reproductive outcomes between CMV IgG-seropositive men and those seronegative. CMV did not seem to play a key role in male reproductive function, as well as in influencing sperm fertility potential in the assisted reproductive outcomes.

KEYWORDS

assisted reproduction, cytomegalovirus, fertility, semen analysis

1 | INTRODUCTION

Cytomegalovirus (CMV) infection is a widespread condition in humans, reaching a prevalence of 50%–80% in adults from central Europe or North America (Onorato, Morens, Martone, & Stansfield, 1985). Since the sexual intercourse does not seem to be a frequent mode of transmission, the oral and respiratory spreads during childhood and probably adulthood appear to be the major way. Following the primary stage of infection, the patients hold the virus in a quiescent state in various tissue cell types for the rest of their life, with intermittent secretions, such as urine, blood, faeces, tears, saliva, breast milk or cervical mucus (Sinzger, Plachter, GreRe, The, & Jahn, 1996). Furthermore, previous studies showed that CMV can be detected also in semen samples, up to 32.7% of men, constituting a reservoir for transmission, although the overall prevalence of CMV DNA finding in seminal fluid seems to be widely variable (Levy et al., 1997; Pass, Stagno, Myers, & Alford, 1980).

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After primary contamination with CMV, virus-neutralising antibodies of immunoglobulin G (IgG) are produced, appearing after 2–5 months, with a complex cell-mediated immune response (Drew & Bates, 1999; Revello & Gerna, 2002).

The latent existence of CMV in the genital tracts, the reactivation of endogenous virus or inter-reinfection between couples, probably can be responsible of viral DNA detection in semen (Shen, Chang, Lin, et al., 1994; Shen, Chang, Yang, et al., 1994).

However, to our knowledge, the progress of CMV infection, the phases of quiescence and reactivation, and the concerns about vertical and horizontal diffusion still remain highly difficult.

Infertility is a main social scourge of the current medicine, as it affects almost 20% of couples in the reproductive age. Nowadays, thanks to the assisted reproductive technologies (ARTs), including in vitro fertilisation/intracytoplasmic sperm injection (IVF/ICSI), couples with severe infertility factors succeed in achieving own biological live births.

The cause of couple infertility may be attributed to the male factor in nearly 40%–50% of cases (Dawson & Whitfield, 1996; Mosher, 1985). However, since in many instances the cause of male factor infertility remains unexplained, searching for new reasons causing male infertility is constantly underway. In this context, the role of viral chronic infections, such as hepatitis immunodeficiency virus (HIV), hepatitis C virus (HCV), hepatitis B virus (HBV) on male reproductive function has been explored (Cito, Coccia, Fucci, Picone, Cocci, Russo, et al., 2019; Cito, Coccia, Fucci, Picone, Cocci, Sessa, et al., 2019; Prisant et al., 2010).

As far as CMV, actually, the attention is mainly focused on the consequences of the prenatally transmission from mother to foetus that can lead to severe disturbances of development, including hearing loss and mental retardation (Levy et al., 1997; Pass et al., 1980). However, the influence of viral infection on the male reproductive health has been received little consideration, despite a potential negative effect of the virus on the spermatogenesis, sperm-mucus interaction and immunologic factors. Moreover, the influence of male CMV on fertility outcomes for assisted reproductive program has never been assessed.

Thus, the aim of this study is to investigate whether the chronic CMV infection can affect semen parameters in men attending ARTs for couple infertility. Secondary objective is to assess the impact of male CMV infection on the reproductive and pregnancy outcomes in CMV-seronegative women suffering from tubal factor infertility.

2 | MATERIALS AND METHODS

2.1 | Study sample

From February 2016 to January 2019, all the couples referring to our ARTs Centre for infertility due to female tubal factor (n = 222) were retrospectively reviewed. The recruitment process methods are shown in Figure 1. From our data record, we selected couples whose men were CMV IgG seropositive (n = 115) and couples whose men were CMV IgG seronegative (n = 107). At the baseline visit, both men and women were routinely screened for serology, according to European guidelines (National Collaborating Centre for Women's and Children's Health (UK), 2013), including HIV 1/2, hepatitis C virus antibody (HCVab), hepatitis B surface antigen (HbsAg), hepatitis B surface antibody (HbsAb), hepatitis B c antibody (HBcAb), Treponema Pallidum Hemagglutination and Venereal Disease Research Laboratories (TPHA-VDRL), Ab anti-Clamidya Trachomatis, CMV IgM and IgG. A previous CMV infection was assessed as positivity for serology to CMV IgG.

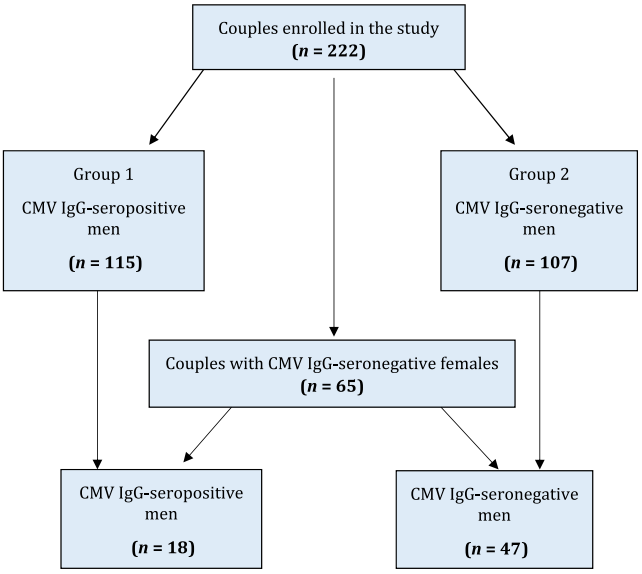

FIGURE 1 Recruitment process

Criteria for inclusion were as follows: (a) CMV IgG seropositive men, aged between 18 and 45 years; (b) CMV IgG seronegative normo-ovulatory women with tubal factor infertility, aged between 18 and 40 years. All females had no evidence of other gynaecological diseases.

We excluded from the analysis men with azoospermia or severe criptozoospermia, cycles using sperm donors, both men and women seropositive for HIV, HCV, HBsAg and CMV IgM.

Moreover, women presenting with diminished ovarian reserve, not adequate response to stimulation protocol, chromosome aberrations or unsuccessful IVF cycles, were omitted from the study. Medical anamnesis and physical examination were performed on both partners.

At baseline, semen analysis was done, by following the 2010 World Health Organization (WHO) guidelines. On the day of fresh IVF/ICSI cycle, seminal parameters were recorded: volume, pH, total sperm count/ml, total sperm concentration, viability, progressive motility (PR), nonprogressive motility (NP), immobility, total motility and normal morphology. For the determination of antisperm antibodies in semen, the mixed antiglobulin reaction (MAR) test was used as described elsewhere (Eggert-Kruse et al., 1991). A MAR test ≥30% was considered positive. After the sperm preparation techniques, we analysed total sperm count/ml, PR motility and NP motility.

All the men were distributed in two groups: Group 1 included CMV IgG-seropositive men, while Group 2 comprised CMV IgG-seronegative patients.

2.2 | Sperm processing

Sperm samples were achieved by masturbation after 2–7 days of sexual abstinence and places into sterile containers, where it was left at 37°C for about 20 min for the complete liquefaction. After liquefaction, ejaculates were examined and categorised (WHO, 2010). Semen samples from CMV IgG-seropositive and seronegative patients were treated under sterility using the density gradient centrifugation technique with 95%, 70% and 50% gradient layers (PureSperm®100, Nidacon). 1–1.5 ml of sample was gradually pipetted on top of the 50% layer and centrifuged at 600 *g* for 10 min; cautiously the pellet was recovered at the bottom of the 95% layer and resuspended in 1.0 ml of warm Flushing Medium (Flushing, Origio, Cooper Surgical Fertility & Genomic Solutions). After centrifugation of the sample at 200 *g* for 10 min, the supernatant was stripped and the pellet was resuspended in 1 ml of fresh medium and was centrifuged again for 10 min at 200 *g*. Finally, the supernatant was taken out and resuspended in a quantity of Fertilization Medium (Fert, Origio, Cooper Surgical Fertility & Genomic Solutions) depending on the sperm concentration. Total sperm count and motility were then evaluated. This pellet was stored in the incubator until the time of insemination.

2.3 | Reproductive outcomes

Therefore, we analysed a subgroup of men, partners of CMV IgG-seronegative normo-ovulatory females, who underwent fresh IVF or ICSI cycles. From here (Figure 1), we selected couples whose men were CMV IgG seropositive (n = 18) and couples whose men were CMV IgG seronegative (n = 47). We recorded data, as follows: aspirated oocytes, inseminated oocytes, fertilised oocytes 2 pro-nuclear (2PN), total fertilised oocytes 1-2-3PN, embryos obtained and embryos transferred. The 2PN fertilization rate (FR), 1-2-3PN FR, cleavage rate (CR), pregnancy rate (PR), miscarriage rate and live birth rate were calculated.

Total and normal oocyte FR were obtained by total number of fertilised oocytes (1-2-3PN) and 2PN fertilised oocytes by the number of injected oocytes respectively. The CR was considered by the number of embryos obtained by the number of 2PN fertilised oocytes.

The luteal phase was provided by a daily administration of 400 mg transvaginal micronised progesterone (Progeffik; Effik) and 50 mg subcutaneous natural progesterone (Pleyris, IBSA) since the day of oocytes retrieval. Embryo transfer (ET) was performed on day 3–5 after IVF/ICSI procedure, using a Wallace catheter (© COOK Medical Incorporated). Supernumerary embryos were frozen, according to our laboratory policy. After 14 days, the human chorionic gonadotropin (HCG) test was done. The clinical pregnancy rate (PR) per cycle and per transfer was calculated by cycles and transfers, respectively, with HCG levels above 50 mU/L and confirmed by transvaginal ultrasounds revealing an intrauterine gestational sac with a heartbeat at around 5–6 weeks of gestation. Pregnancy loss before 20 weeks of gestation and all biochemical pregnancies were contemplated as miscarriages. Live birth rate per cycle was defined as the percentage of all cycles and transfers, respectively, that lead to live births.

2.4 | Ethics and statistics

All the patients gave a written informed consent to agree on having their data included into the study. All procedures were performed in accordance with the ethical standards of the institutional and national research committee and with the 1975 Helsinki Declaration. Continuous variables are presented as the median and interquartile range (IQR). Differences between groups were assessed using a Kruskal–Wallis or Mann–Whitney *U* test as appropriate. Categorical variables were tested using a χ^2 test or Fisher's exact test. A p-value < .05 was set as statistically significant. All collected data were evaluated with Statistical Package for Statistical Sciences (SPSS, version 22.0, IBM).

3 | RESULTS

This retrospective single-centre cohort analysis included 222 men, partners of women suffering from tubal factor infertility. One hundred and fifteen (51.8%) CMV IgG-seropositive men were included in the Group 1, 107 (48.2%) CMV IgG-seronegative men were comprised in the Group 2.

The median male age was 39 ± 4.9 years old (IQR: 36–42). Baseline characteristics of the semen analysis were showed in Table 1. There was reported a low trend towards higher sperm concentration/ml, total sperm count and viability in CMV IgG-seronegative males, compared to CMV IgG-seropositive patients. However, no statistically differences were found among groups (p = .42, .60 and .87 respectively). As depicted in Table 2, data of sperm volume, pH, motility PR, motility NP, immobility, total motility and normal sperm morphology were similar between the two groups, without reaching statistically significance (p > .05). Also after the sperm preparation techniques, the semen parameters of CMV IgG-seropositive men remained comparable to those of CMV IgG-seronegative men. The MAR test was negative in all patients. Leukocytospermia has not been detected under any circumstances.

Considering the subgroup of men, partners of CMV IgG-seronegative females, 65 couples (29.2%) were selected for the analysis (Table 3). Overall, 65 fresh cycles were performed: 29 (44.6%) women underwent IVF and 36 (55.4%) cycles were done by ICSI. Eighteen couples (27.7%) were serodiscordant for serology (detection of IgG antibodies to CMV in male serum only and

TABLE 1 Baseline characteristics of the semen analysis before and after sperm preparation techniques (*N°* = 222)

Variable	Median (IQR)
Male age, years	39 (36–42)
CMV IgG-seropositive men, *n* (%)	
No	107 (48.2)
Yes	115 (51.8)
Semen volume, ml	3.1 (2.2–3.95)
Semen pH	7.70 (7.60.–7.80)
Sperm concentration, million/ml	31 (11–53)
Total sperm count, million/ejaculate	91 (24–176.4)
Viability, %	72 (67.25–80)
Progressive motility (PR), %	50 (39–60)
Nonprogressive motility(NP), %	10 (5–10)
Immotility, %	40 (30–50)
Total Motility, %	60 (50–70)
Normal morphology, %	5 (4–6)
After sperm preparation techniques	
Sperm concentration, million/ml	4.5 (1.6–10)
Progressive motility (PR), %	90 (80–95)
Immotility, %	10 (5–20)
Total motility, %	90 (80–95)

not in female serum). No case of seroconversion to the partner was recorded. The median male age was 39 ± 4.0 years old (IQR: 34–40), and the median female age was 37 ± 4.2 years old (IQR: 33–39). Overall, the median 2PN FR was 67% (IQR: 50%–87%), the median total FR was 83% (IQR: 63%–100%). The median CR was 100% (IQR: 100%–100%). Pregnancy was achieved in 17/65 cycles (PR/cycle = 26.2%). The PR/transfer was 32.0% (17/53). In 7/65 (10.8%) cycles, miscarriages were reported. Live births were obtained in 10/65 cycles (live birth rate/cycle = 15.4%). The live birth rate/transfer resulted 18.9%. Overall, we achieved nine singleton pregnancies and one twin pregnancy.

As shown in Table 4, comparing the 2PN FR, total FR and CR between the group with CMV IgG-seropositive men and the one with seronegative men, no significant differences were found (p = .33, .52 and .08 respectively). Equally, PR/cycle, miscarriage and live birth rates appeared similar between the two groups (p = .54, .63 and .50 respectively).

TABLE 2 Comparison of semen parameters between Group 1 (CMV IgG-seropositive men) and Group 2 (CMV IgG-seronegative men)

Variable	Group 1 (CMV IgG +)	Group 2 (CMV IgG −)	*p* value
Semen volume, ml	3.2 (2.3–4)	2.9 (2–3.7)	.73
Semen pH	7.60 (7.60–7.80)	7.60 (7.60–7.80)	.38
Sperm concentration, million/ml	28 (8.4–50.5)	38 (17–62)	.42
Total sperm count, million/ejaculate	93.2 (22.2–179.5)	100.8 (40.3–196.1)	.60
Viability, %	72 (64.5–80)	73.5 (70.25–80)	.87
Progressive motility (PR), %	50 (35–60)	50 (35–60)	.29
Nonprogressive motility (NP), %	10 (5–10)	10 (5–15)	.31
Immotility, %	40 (30–50)	40 (35–50)	.21
Total motility, %	60 (50–70)	60 (50–65)	.21
Normal morphology, %	5 (4–6)	5 (4–6)	.64
After sperm preparation techniques			
Sperm concentration, million/ml	4 (1.5–10)	6 (2.4–10)	.11
Progressive motility (PR), %	90 (80–95)	90 (80–95)	.17
Immotility, %	5 (5–20)	10 (5–20)	.36
Total motility, %	95 (80–95)	90 (80–95)	.33

Note: Variables are expressed in median and interquartile range (IQR). Statistical significance was considered as $p < .05$.

4 | DISCUSSION

Recently, the influence of chronic viral infections on male fertility has been considerably highlighted. Indeed, several viruses can colonise the genital tracts and semen, generating effects extremely important in terms of organ integrity and changes in the reproductive and endocrine systems. In particular, HIV and HCV infections proved to be able to reduce progressive sperm motility, in comparison with normozoospermic healthy seronegative controls (Cito, Coccia, Fucci, Picone, Cocci, Russo, et al., 2019; Cito, Coccia, Fucci, Picone, Cocci, Sessa, et al., 2019).

Equally, some authors detected that HBV seropositive men have lower sperm motility and total sperm count, as well as poor morphology (Oger et al., 2011; Zhou et al., 2011).

After the primary infection, CMV endures in the human body throughout the lifetime. Indeed, several studies estimated a prevalence of viral DNA in the seminal tracts of fertile and infertile men that range from 8% to 65% (Bresson et al., 2003; Eggert-Kruse, Reuland, Johannsen, Strowitzki, & Schlehofer, 2009; Neofytou, Sourvinos, Asmarianaki, Spandidos, & Makrigiannakis, 2009). In this setting, the impact of the chronic CMV infection on male reproductive health is currently a debated issue.

A previous experimental study, conducted in male mice, investigated the effects of the CMV injection in the testes. As demonstrated, the peritubular cells appeared severely damaged, as well as spermatogenesis altered, but germ cells and Sertoli cells were not involved. Following the mating of infected males with seronegative females, neither infectious virus nor viral DNA was found in spermatozoa retrieved from uterine fluid, demonstrating that

TABLE 3 Subgroup of couples with CMV IgG-seronegative females (*N°* = 65)

Variable	Value
Male age, years	39 (34–40)
Female age, years	37 (33–39)
CMV IgG-seropositive men, *n* (%)	
No	47 (7.2)
Yes	18 (27.7)
IVF cycles, *n* (%)	29 (44.6)
ICSI cycles, *n* (%)	36 (55.4)
Oocytes retrieved, *n*	6 (4.5–8.5)
Oocytes inseminated, *n*	5 (3.5–8)
Oocytes 2PN fertilised, *n*	4 (2–6)
Oocytes total 1-2-3PN fertilised, *n*	4(2–7)
Fertilisation rate 2PN, %	67 (50–87)
Fertilisation rate 1-2-3PN, *n* median (IQR)	83 (63–100)
Cleavage rate, *n*	100 (100–100)
Embryos obtained, *n*	3 (2–5.5)
Embryos transferred, *n*	2 (1–2)
Pregnancy rate/cycle, *n* (%)	17 (26.2)
Pregnancy rate/transfer, *n* (%)	17 (32.0)
Miscarriage rate/cycle, *n* (%)	7 (10.8)
Miscarriage rate/transfer, *n* (%)	7 (13.2)
Live birth rate/cycle, *n* (%)	10 (15.4)
Live birth rate/transfer, *n* (%)	10 (18.9)

Note: Variables are expressed in median and interquartile range (IQR).
Abbreviations: ICSI, intracytoplasmic sperm injection; IVF, in vitro fertilisation; PN, pronuclear.

TABLE 4 Comparison of reproductive outcomes between CMV IgG-seropositive and CMV IgG-seronegative men, considering cycles with CMV IgG-seronegative women (*N°* = 65)

Variable	CMV IgG +	CMV IgG −	*p* value
Oocytes retrieved, *n*	6 (5.75–8.5)	6 (3–9)	.45
Oocytes inseminated, *n*	6 (4–8.25)	5 (3–8)	.33
Oocytes 2PN fertilised, *n*	4.5 (2–6.25)	3 (2–6)	.48
Oocytes total 1-2-3PN fertilised, *n*	4.5 (2.75–7)	4 (2–7)	.45
Fertilisation rate 2PN, %	0.81 (0.5–0.92)	0.67 (0.5–0.83)	.33
Fertilisation rate 1-2-3PN, *n* median (IQR)	0.85 (0.69–1)	0.80 (0.60–1)	.52
Embryos obtained, *n*	4 (2–6.25)	3 (2–5)	.45
Embryos transferred, *n*	2 (1–2)	2 (1–2)	.84
Cleavage rate, *n*	1 (1–1)	1 (0.90–1)	.08
Pregnancy rate/cycle, *n* (%)	5 (27.8)	12 (25.5)	.54
Miscarriage rate/cycle, *n* (%)	2 (11.1)	5 (10.6)	.63
Live birth rate/cycle, *n* (%)	3 (17.6)	6 (14)	.50

Note: Variables are expressed in median and interquartile range (IQR). Statistical significance was considered as $p < .05$.
Abbreviation: PN, pronuclear.

CMV latent in the male genital organs was not transferred to the offspring, even during the acute stage of CMV disease (Tebourbi et al., 2001).

Regarding the human studies, most of them agreed on finding no association between CMV infection and male fertility (Bezold et al., 2007; Eggert-Kruse et al., 2009; McGowan, Hayes, Kovacs, & Leydon, 1983; Neofytou et al., 2009; Pallier et al., 2002), while others revealed that sperm parameters and functions are injured by the virus (Klimova et al., 2010; Wu et al., 2007), and that high amounts of CMV in the semen are related with decreased percentages of sperm motility (Lang, Kummer, & Hartley, 1974). This probably might occur because patients with CMV in the ejaculates suffer from simultaneous chronic inflammatory urogenital tract diseases that may reduce total sperm quality (Naumenko et al., 2014). In this context, all patients enrolled in our study did not report history of recent inflammatory urogenital disorders, nor presented leukocytospermia in the seminal fluid.

In a study on HIV patients, CMV was identified in semen CD45+ cells, but not in mature spermatozoa (Rasmussen, Morris, Hamed, & Merigan, 1995), while a more recent study on CMV-infected organotypic cultures explained the lytic effects of viral antigens and viral particles in spermatogonia, spermatocytes and spermatids. These infected cells could develop to mature CMV-carrying spermatozoa, but mature spermatozoa did not appear to sustain the infection (Naumenko et al., 2011). Thus, a significant decrease in the number of immature germ cells suggested that CMV has a direct gametotoxic effect, contributing to infertility.

Otherwise, CMV in semen seems to not significantly impact on semen quality (Bantel-Schaal, Neumann-Haefelin, & Schieferstein, 1993; Eggert-Kruse et al., 2009; McGowan et al., 1983; Shen, Chang, Lin, et al., 1994; Shen, Chang, Yang, et al., 1994; Yang et al., 1995). In a previous study, the experimental viral contamination of ejaculated spermatozoon in vitro was achieved, as well as the adherence of viral particles to the sperm membrane was confirmed, although no effects on sperm motility were observed (Pallieret al., 2002). Indeed, CMV DNA seemed to be able to attach the surface of spermatozoa and to enter in these cells through the intact plasma membrane. However, albeit CMV has been proven to be present in a large amount of ejaculates, no relationship with the outcomes of semen was found in many studies (Eggert-Kruse et al., 2009).

These findings confirm our data and strengthen our theory that human CMV does not seem to be a substantial cause of male infertility. Indeed, no relationship between CMV and sperm quality declining was found in our study.

Furthermore, the washing procedure utilised for ARTs is estimated to remove viral particles or decrease viral load, although the risk of viral diffusion cannot be absolutely ignored, especially when ICSI is performed (Pallier et al., 2002). In our case series of serodiscordant couples (male seropositive and female negative), independently of the ARTs technique used, seroconversion of the female partner did not occur under any circumstances.

Based on the lack of association between serological findings and evidence of CMV in the male genital tract, we believed that serology is adequate to screen for CMV infection.

Indeed, CMV IgG and IgM represent easy tools for choosing patients who are at risk of cytomegalovirus infection. In this regard, the prevalence of CMV IgG patients in our study (51.8%) reflected the findings of other studies.

Moreover, the effects of human CMV on cellular metabolism may also generate effects on the activation or replication of other viruses in co-infected cells, which might impact on sperm function (Rasmussen et al., 1995). Nevertheless, no other viral infections, including HIV, HCV or HBV, were detected in our study population.

As regards the influence of chronic viral infections on the ARTs outcomes, previous studies demonstrated that HIV or HCV are able to reduce fertilisation rates in a group of serodiscordant couples, with seropositive male and negative female (Bantel-Schaal et al., 1993; Cito, Coccia, Fucci, Picone, Cocci, Russo, et al., 2019; Cito, Coccia, Fucci, Picone, Cocci, Sessa, et al., 2019). This possibly happens as a result of the antioxidant defences declining, including superoxide dismutase and glutathione peroxidase (Levent et al., 2006; Salemi et al., 2010). However, the overall pregnancy and live birth rates in HIV and HCV-seropositive men were comparable to those of the matched control seronegative group.

Likewise, as shown by a recent study, the presence of HBsAg in serum of men belonging to serodiscordant couples has been demonstrated to affect significantly total sperm fertilisation capacity, thus reducing the total FR (Cito, Coccia, Fucci, Picone, Cocci, Russo, et al., 2019; Cito, Coccia, Fucci, Picone, Cocci, Sessa, et al., 2019).

On these bases, we went to evaluate even the function of CMV infection and we found that fertilisation, as well as pregnancy outcomes of CMV IgG-positive men, was similar to those of seronegative men.

To the best of our knowledge, this is the first study that aims to assess the impact of chronic CMV infection on sperm fertilising potential, considering the reproductive outcomes of an assisted reproductive program. For this purpose, we included in the study, women with only tubal infertility, in order to remove all possible bias related to the female factor. However, we did not test the presence of CMV DNA in the seminal samples, due to the time and cost of performing polymerase chain reaction (PCR) and cell culture, compared to the more rapid and inexpensive detection of CMV IgG and IgM assays.

5 | CONCLUSION

In all couples attending an ARTs Center, serologic tests for the detection of previous cytomegalovirus infection are recommended. The viral infection did not seem to play a key role in male reproductive function, as well as in influencing sperm fertility potential in the assisted reproductive outcomes. However, CMV serology screening could be crucial to identify primary acute infection by detecting IgM seropositivity, in order to prevent virus transmission by sperm sample.

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