TORCH, Anti-cardiolepin, and Anti-phospholipids in Women with Repeated Miscarriage in Mosul City

Karam A. Aldabbagh, Mohammed KJ. Alnori, Khalid W. Almola

1Department of Clinical Laboratory Sciences, College of Pharmacy, University of Mosul, Mosul, Iraq; 2Department of Microbiology, College of Medicine, University of Mosul, Mosul, Iraq

Abstract

BACKGROUND AND OBJECTIVE: The present study aimed to identify a connection between common viral infections and women who had repeated miscarriages.

MATERIAL AND METHODS: This study is a retrospective study which has been carried out on 2010 records in Mosul city/Iraq, and the patients were all women of child-bearing age (20–35 years old). All of the women had suffered from an abortion at least once before. The target viruses in charge for this study include Toxoplasma gundi, Cytomegalovirus, Rubella virus, and Herpes virus. The patient’s records include tests for detection of serum antiphospholipid, and anticardiolepin, together with cutoff values of Toxoplasma gundi, Cytomegalovirus, Rubella virus, and Herpesvirus as a positive result for the presence of these infectious diseases.

RESULTS: The analysis is based on the detection of the antibodies which confirmed a negative correlation between antibodies level and active viral infection, together with no clear association between the presence of active infectious status and miscarriage. In addition, only a few positive cases for each patient were positive for viral infection.

CONCLUSION: The outcome confirmed no clear association between the presence of infectious agents and repeated miscarriage. Nonetheless, rubella and cytomegalovirus have shown the highest contribution in this regard.

Introduction

Miscarriage is officially defined as the loss of pregnancy within the first 12 weeks. Late miscarriage is defined as the loss of pregnancy from 12 to 24 weeks [1], [2]. It is fairly common since it occurs in one of five pregnancies. It can have considerable physiological and psychological implications for the patient [3], [4]. Infection can cause nearly 15% of early miscarriages and 66% of all later miscarriages [5]. Reports increasingly accumulating regarding the association between abortion and the presence of viral infections; including Herpes virus [6], [7], human papillomavirus [8], [9], Parvoviruses [10], Cytomegalovirus [8], [9], Rubella virus [11], [12], [13], or protozoal infection, such as Toxoplasma Gondi [14], [15] and bacterial infection [16].

Anti-cardiolipins are antibodies that are directed against cardiolipin which is found in several diseases including syphilis and anti-phospholipid syndrome. The presence of these antibodies in a pregnant woman’s body could result in spontaneous abortion or miscarriages. Anti-phospholipid syndrome is a disorder of the immune system which can cause an increased risk of blood clots within the body. This means that those with this condition are at greater risk of developing deep vein thrombosis which results in blood clots in the leg. This has also been linked to pregnancy complications and recurrent miscarriages. It can also result in late miscarriage which is caused after 12 weeks of pregnancy [17], [18], [19], [20].

Although abortion seems to be the most prevalent unfavorable pregnancy result, global statistics are unavailable. Hence, it is a very significant factor to watch out for when handling the treatment and care of women who have miscarried before. Several publications have associated anti-phospholipid syndrome with retarded intrauterine growth and others have associated it with pre-eclampsia. The present study focuses on analyzing the prevalence of miscarriage in pregnancy in our locality since such statistics are lacking.

Patients and Methods

This study is a retrospective study where data were collected from the main gynecological department...
in the city hospital. Records of all miscarriage mothers were considered regarding the presence of viral infection. The patients were all women of childbearing age (20–35 years old) and suffered from abortion at least once before. No interference was conducted to select patients’ data, the included data were kept in the same order as they were conducted and recorded by the laboratory staff.

The included tests were the measurement of anti-Toxoplasma Gondi IgM antibodies and the measurement of anti-HSV IgM antibodies in a patient’s serum by ELISA as referred by the specialized obstetrician; IgM antibodies represent the marker of active infection rather than IgG antibodies which represent an old or latent infection. All ELISA kits were from bioMerieux Company and the ELISA BioTek ELX-800 instrument was used as instructed by the kit insert. The accuracy rate of anti-Toxoplasma Gondi IgM antibodies and anti-HSV IgM antibodies kits is 97.4% and 97%, respectively [12], [13], [14], [15], [16], [17], [18], [19], [20]. Routine tests for miscarriage women also include detection of serum concentration of anti-cardiolipin and antiphospholipid as a confirmative technique for the presence of infectious diseases including these mentioned viruses.

Statistical analysis in this study was conducted using Microsoft Excel spreadsheets version 14.0. 2010, using a T-test at p ≤ 0.05 was considered significant.

**Results**

**Correlation between infectious agent and measured antibody parameters**

The study found that there were no significant instances of anti-phospholipid and anti-cardiolipin within the patients who had either of the four viruses. This showed that since the antibodies were absent, this meant that the patients had been infected. As a result, these viruses likely affected repeated miscarriages within the few correlated patients. The lowest occurrences of the antibodies were within patients who had either the Rubella or the Herpes virus. Not only are these common viruses, but are known to be explicitly deadly against unborn babies. Table 1 results show the correlation between infectious agents such as Toxoplasma Gondi, Cytomegalovirus, Rubella Virus, and Herpes Virus. This table has also defined a set of values to understand the data more precisely.

<table>
<thead>
<tr>
<th>Correlation</th>
<th>Anti-phospholipid</th>
<th>Anti-cardiolipin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Toxoplasma Gondi</td>
<td>0.16</td>
<td>0.033</td>
</tr>
<tr>
<td>Cytomegalovirus</td>
<td>0.018</td>
<td>0.01</td>
</tr>
<tr>
<td>Rubella Virus</td>
<td>0.01</td>
<td>0.001</td>
</tr>
<tr>
<td>Herpes Virus</td>
<td>0.02</td>
<td>0.015</td>
</tr>
</tbody>
</table>

**Discussion**

Herpes is viral DNA and investigators have confirmed that this virus enters the host and remains dormant lifelong with potential reactivation [21]. In a study...
conducted by Kapranos and Kotronias, 2009 [6], using DNA hybridization of sera samples of women undergoing recurrent spontaneous abortion compared to a lower rate of elective abortion. Similarly, Kim et al., 2012b reported that an analysis of sera of pregnant women for HSV found a relatively high positivity (17%) [7]. Rubella has been partially studied and inconclusive outcomes reported by researchers. Rubella has coexisted with other virus infections such as HSV [7].

In a study conducted by Hadar et al., 2010 who has tested a group of women with CMV positive before pregnancy [22]. Only four out of 59 women enrolled in the study had miscarriage other underwent normal delivery or elected abortion making the outcome inconclusive. These controversial results were recorded in another more detailed study conducted by Saraswathy et al. 2011 who has shown the presence of CMV IgG antibody in their serum [23].

The protozoal infection has been increasingly linked to miscarriage. Toxoplasmosis has been investigated in all miscarriages studied population. In a study conducted on 100 serum samples from 55% toxoplasma seropositive subjects, most of them were in the first trimester, however, the study lacked comparison to the control group [24]. Similarly, Alvarado-Esquivel et al. 2014, study lacked comparison to control and confirmed a slightly high exposure to toxoplasma Gondi among miscarriage women [25]. In alternative meta-analysis studies conducted over 132 studies, the outcome has shown a higher infection rate among miscarriage women [26]. Nonetheless, epidemiological studies have confirmed that the link between Toxoplasma infection rate and miscarriage differs across the world and varies from 40 % to 70% between UK/USA and tropical countries, respectively [27].

The immune system’s participation in a good pregnancy is critical. While the semi-allogeneic fetus’ immunological responsiveness is established, various components of the immune system perform their assigned duties in preparation for implantation as well as during gestation [28], [29]. In the fetomaternal interface, natural killer (NK) cells, macrophages, and dendritic cells have all been found [30], [31], [32]. Cytokines such as interleukin (IL-10) and colony-stimulating factor (CSF-1) have been connected to the implantation process and are present in uterine cells [30], [33]. Due to the obvious infiltration and injury to the maternal tissue, implantation causes an inflammatory reaction, with several cells suffering apoptosis [34], [35].

In contrast, inflammatory cytokines like interferon-g and tumor necrosis factor-alpha (TNF-a) are not normally produced in the placenta and have been linked to miscarriage in mice models [28]. Stillbirth is hypothesized to be caused by abnormal implantation, placentation, or blood vessel change [4], [36]. An active infection might disturb the pregnancy by interfering with any of the above-mentioned processes as well as disrupting the immunological homeostasis, even if it never culminated in maternal and fetal infection.

The mechanism of maternal-fetal transfer of infectious agents has been studied carefully. Some of these infectious agents are transferred to the fetus through maternal circulation, such as plasmodium Malaria [37], [38]. Other infectious agents are using surface protein to internalize into the placenta such as Listeria monocytogenes [39], [40], [41] using invasion mechanism of internalization through placenta.

Since the cause of miscarriage is yet unknown, many factors have been regarded as contributing to the increase in the chance of miscarriage, these include genetic, mother stress and psychological status, mother’s high/low BMI, mother malnutritional, alcoholics, smoking, and use of NSAIDs [2], [42], [43], [44], [45], for these aforementioned causes, sequential miscarriage is considered in women with previous history of first miscarriage [46]. Finally, reports also indicated the reciprocal link to presence of infectious agents [47], [48], [49], [50], [51], [52]. However, no single infectious agent was conclusively agreed to be responsible, moreover, bacterial, viral, and protozoal
are believed to be equally involved, thereby, no single infectious agent could be ruled out [5]

### Conclusion

The data analysis of this study has shown that while there was a significant presence of each of the four viruses mentioned within the sample population of female patients, none could be singled out as a deciding factor for repeated miscarriages. The statistical truth wills out in this study, that 60% of all miscarriages are unexplained; however, many can be explained by the occurrence of infectious diseases or the presence of infectious microorganisms; with rubella and cytomegalovirus seeming to be the main causative agents in this regard. Underestimation of miscarriage's physical and psychological number of casualties are common. The direction from diagnosis through professional management to supportive aftercare can be difficult. The rate of robust toxoplasmosis and HSV infectivity in frequently aborted females shows no link to weather changes. The active HSV infectivity is more associated with abortion than toxoplasmosis in this set of experienced females. Varying terms and inclusion criteria hamper comparison of treatment options for miscarriage in studies, multiple doses and routes of administration of drug treatment, and different definitions of treatment failure. Other infectious diseases need to be aligned together and further studies need to be assessed.

We do recommend routine screening tests to be conducted in our local teaching hospitals to identify positive cases if any. These screening tests will improve the follow-up for those positive cases which might reduce the rate of continuous abortion in this context.

### Acknowledgments

The authors are very grateful for the staff in gynecological department in the city hospital, thanks are also for the college of pharmacy/University of Mosul for their help to complete this work.

### Authors Contributions


### References

Aldabbagh et al. Role of TORCH in Miscarriage


975

rsf.2010.0320
PMid:20659931

PMid:24520948


PMid:24093517

PMid:31573753

PMid:11311759

PMid:20795446

PMid:22555404


21. Whitley RJ, Roizman B. Herpes simplex virus infections. PMid:28985591


PMid:11432815

PMid:15073336

PMid:18806773

PMid:1752937

PMid:11157526

PMid:12033743

PMid:15142995

PMid:16428886

PMid:20618237

PMid:15590426

PMid:14526331

PMid:12385841

PMid:16949414

PMid:18192789