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Oxidative Stress: A Hidden Factor in Miscarriage

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ABSTRACT

Background: *Miscarriage or spontaneous pregnancy loss is the unintentional termination of a pregnancy before fetal viability at 20 weeks of gestation or when fetal weight is <500 grams. Oxidative stress represents an imbalance between the generation and manifestation of free radicals or reactive oxygen species (ROSs) and ability of the biological system to readily detoxify the reactive middle or to repair the consequential damage. Oxidative stress of placenta, embryo and fetus plays an important role in pathogenesis of many pregnancy complications including miscarriages, preeclampsia and preterm labour.*

Objectives: *To estimate oxidative stress in women with miscarriage and compare it with controls and to establish oxidative stress (MDA) as a clinical marker for miscarriage and SOD levels as an indicator for safe pregnancy.*

Materials and Methods: *70 women with miscarriage (study group), and 70 with healthy pregnancy (control group) were enrolled. Serum MDA and serum SOD levels were compared in total, 1st trimester and 2nd trimester study and control group as well as in 1st, 2nd and 3rd miscarriage females prior to 24 weeks of gestational age.*

Results: *Serum MDA levels were found to be significantly higher while serum SOD levels were found to be significantly lower in total (p= <0.0001, 0.0016), 1st trimester (p= <0.0001, 0.0085) and 2nd trimester (p= <0.0001, 0.0431) study group on comparison with control group. Moreover, there was increase in oxidative stress with each miscarriage shown by enhanced MDA and diminished SOD levels in women with repeated miscarriages.*

Conclusion: *Present deliberation manifests that hoisted serum MDA and abridged serum SOD levels might be involved in the pregnancy loss and ostracism of fetoplacental material out of the uterine cavity and may play an imperative role in the etiology of spontaneous abortion.*

Keywords: *Malondialdehyde, Miscarriage, Oxidative stress, Superoxide dismutase*

I. INTRODUCTION

Reproduction in humans is marred by early pregnancy failure. Miscarriage is also known as spontaneous pregnancy loss (SPL) or spontaneous abortion (SA) is the termination of pregnancy before fetal viability at 20 weeks of gestation or delivery of all or any part of the products of conception, with or without a fetus weighing less than 500 grams [1, 2]. The recognized etiological factors may be either alterations in maternal environment in which the fetus grows involving infections, anatomical, endocrinological, genetical and immunological

abnormalities or embryonic defects like chromosomal abnormalities which inhibit the development of embryo. In addition, many other causes are related to environment and life style like cigarette smoking, obesity, cocaine use, alcohol and increased caffeine consumption [3]. Moreover, the incidence of miscarriage is influenced by the age of the mother and a number of pregnancy related factors, including a history of a previous full-term normal pregnancy, number of previous spontaneous abortions and a previous stillbirth [2].

Oxidative stress occurs due to increased generation and manifestation of free radicals or reactive oxygen species (ROSs) and diminished ability of the biological system to readily detoxify the reactive middle or to repair the consequential damage. OS occurs when the generation of ROSs and other radical species exceeds the scavenging by antioxidants as a result of excessive production of ROSs, inadequate intakes or increased utilization of antioxidants [4].

Oxidative stress, despite the way its name suggested, is a natural phenomenon in the body. However, for the past two decades or so, OS has gained much importance in the field of research due to its association with multiple pathological conditions. In our body system, ROSs or free radicals are continuously formed as a consequence of biochemical reactions, e.g. within the mitochondrial respiratory chain and from external factors. To cope with the ROSs, animal and human cells have developed a ubiquitous antioxidant defense system, which consists of enzymatic and non enzymatic antioxidants. Whenever the natural balance between the rate of generation of free radicals and action of antioxidant is lost, it results in OS [5]; which in turn modulate cellular functions and impair the intracellular milieu, resulting in diseased cells or endangered cell survival [6] by acting on lipids and proteins [7]. Irreversible modification of cellular components by ROSs leads to cell dysfunction, leading to functional alteration and even cell death as shown in Fig. 1.

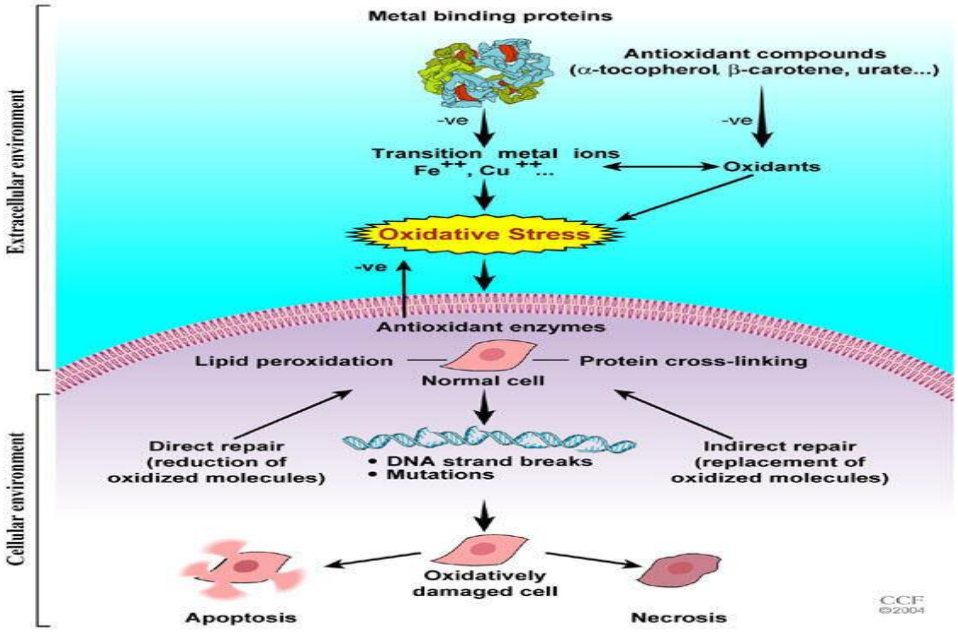

Figure 1. Mechanisms of oxidative stress-induced cell damage [6].

In pregnancy, increased oxygen (O_2) demand makes the body vulnerable to ROSs formation, particularly within the mitochondria due to leakage of electrons from enzymes of the respiratory chain. The resulting stress leads to impaired placental development and degeneration of the syncytiotrophoblast. This might be because of location of syncytiotrophoblast on the villous surface and much lower concentrations of the antioxidant enzymes during early gestation than other villous tissues [8, 9]. As OS has a pathophysiologic role in the placenta, embryo, and the fetus [10]; increased free radicals activity is seen in women with RSA [11] and may result in other complications like spontaneous abortion, preeclampsia, and intrauterine growth restriction (IUGR) [12].

So as far the reports, the key factors responsible for miscarriage are multiple ranging from genetic, structural, infectious, immunological, metabolic, endocrine and environmental. Most of the studies regarding oxidative stress have been conducted in women with recurrent spontaneous abortion, while the data regarding their role in single pregnancy loss is inadequate. The present study may be a step towards trimming down the diagnostic window to identify the oxidative stress contributory factors for miscarriage at early stage as well to relieve the agony and suffering of the couples as a result of repeated loss of desirable pregnancies. Therefore, the present study has been planned with following objectives:

1. To estimate oxidative stress in women with miscarriage and compare it with controls.
2. To establish oxidative stress (MDA) as a clinical marker for miscarriage and SOD levels as an indicator for safe pregnancy.

II. MATERIALS AND METHODS

The present study was conducted on 70 clinically confirmed cases of miscarriage attending the OPD and indoor of Department of Obstetrics and Gynecology, Adesh Institute of Medical Sciences and Research, Bathinda and from Civil Hospital, Bathinda. For comparison 70 females with healthy ongoing pregnancy constituted the control group. Both the participating groups were age and gestational age matched. We further stratified study population into 1st and 2nd trimester to confirm any difference in demographic, anthropometric and biochemical parameters and into 1st, 2nd and 3rd miscarriage; again to confirm if there was augmented deterioration in SOD and enhancement in MDA levels with repeated miscarriage.

Study protocol

All the enrolled cases were subjected to demographic, anthropometric and biochemical analysis. A detailed history with full clinical examination was performed in all the cases. Also, the information about socio economic status, personal history like hypertension/diabetes mellitus/smoking/alcohol/drug abuse was taken. An informed written consent was obtained from the study participants in their own language prior to enrollment in the study.

Selection criteria: Inclusion criteria:

Study group: Clinically confirmed cases of miscarriage

Control group: Females with healthy ongoing pregnancy with no previously recognized miscarriage; no clinical evidence of endocrine abnormality constituted the control group.

Exclusion criteria: Patients with history of thyroid dysfunction, diabetes mellitus, PCOS, heart disease, hypertension, uterine fibroid, infectious disease, smokers and alcoholics were excluded from the study.

Serum levels of **Malondialdehyde** were estimated by Kei Satoh Method [13].

Serum **Superoxide dismutase** (SOD) was estimated by method of Marklund and Marklund [14].

Statistical analysis

Statistical analysis of the collected data was done by using Microsoft Excel 2007 software and various online calculators. The results of laboratory tests in the study and control groups were summarized as mean ± standard deviation. Comparison between subjects (both participating groups) was done using Student's unpaired *t* test and χ^2 test. 95% confidence interval was taken into consideration and $p < 0.05$ was regarded as statistically significant. Correlation (Pearson) analysis was used to test the linear relationship between MDA and SOD.

III. RESULTS

The present study enrolled maternal age and gestational matched study and control group. Mean maternal age was 26.80±4.19 years in study group and 25.53±3.88 years in control group. Mean gestational age was 12.31±4.68 weeks in study group and 12.87±4.63 in control group.

Depending on cause and stage of pregnancy, the distribution of study group females according to type of spontaneous abortion showed (Fig. 2) that majority of women presented with missed (37%) and incomplete (33%) abortion, while 13% were with inevitable abortion, 10% with complete abortion and 7% with threatened abortion. Furthermore as per distribution of study group females according to occurrence of miscarriage (Fig. 3), 69% women represented with first miscarriage, 27% with second miscarriage and 4% were with third miscarriage.

In order to check whether rural women population is more prone to miscarriage, the comparison of area wise distribution of study and control group population was done, which showed (Table 1) 60% of study and 53% of control group belonging to rural area, while, 40% of study and 47% of control participants were belonging to urban area. On statistical analysis, the difference was found to be non significant (p= 0.3941).

To facilitate if women age at marriage affects the risk of miscarriage, comparison of marriage age of total, 1st trimester and 2nd trimester study and control group females was done (Table 2). Mean age at marriage was 21.44± 3.39 years (range 15-33 years) in total study group and 22.01±3.08 years (range 16-31years) in control group and difference was found to be non significant (p= 0.3016). Mean age at marriage for 1st trimester and 2nd trimester study and control group was 21.83± 3.56 vs. 21.97±2.93 years and 20.90±3.12 vs. 22.07±3.33 years respectively and difference was found to be again non significant (p= 0.8424, 0.1719).

In order to see whether age of women, when they first become pregnant, influence the further risk of miscarriage, comparison of age in study and control group at 1st pregnancy in total, 1st trimester and 2nd trimester was carried out (Table 3). Mean age at 1st pregnancy was 23.37± 3.60 years (range 17-34 years) in study group and 23.50±2.86 years (range 18-32 years) in control group and the difference was found to be non significant (p= 0.8165). The mean age at 1st pregnancy for 1st trimester and 2nd trimester study and control group was

23.73±3.70 vs. 23.38±2.72 years and 22.86±3.44 vs. 23.66±3.09 years respectively and again difference was found to be non significant (p= 0.6334, 0.3594).

BMI in total, 1st trimester and 2nd trimester study and control group was recorded and compared to find if women with higher BMI are more likely to pregnancy loss (Table 4). On comparison, mean BMI was 21.33±4.18 Kg/m^2 (range 12.5-35.41 Kg/m^2) in total study group and 20.12±3.15 Kg/m^2 (range 15.07-29.64 Kg/m^2 years) in control group and difference was found to be non significant (p= 0.0563). Mean BMI for 1st trimester and 2nd trimester study and control group was 21.17±4.37 vs. 19.85±2.91 and 21.54±3.95 vs. 20.48±3.46 Kg/m^2 years respectively and again difference was found to be non significant (p= 0.1110, 0.2803).

To study, if central obesity is a risk factor for miscarriage, comparison of WHR of total, 1st trimester and 2nd trimester study and control group population was made (Table 5). Mean WHR was 0.91±0.04 cm (range 0.80-0.97cm) in total study group and 0.91±0.04 cm (range 0.80-0.97cm) in control group and difference was found to be non significant (p= 0.7553). Mean WHR for 1st trimester and 2nd trimester study and control group was 0.92±0.04 vs. 0.91±0.03 and 0.90±0.04 vs. 0.91±0.04 cm respectively and difference was found to be again non significant between both the participating groups (p= 0.7318, 0.4403).

In order to establish MDA levels as a marker of oxidative stress in women with miscarriage, comparison of MDA levels in total, 1st trimester and 2nd trimester study and control group was done which showed (Table 6) that mean serum MDA levels in total were 7.34±2.02 nmol/ml (range 3.13-13.59 nmol/ml) in the study group and 4.61±1.35 nmol/ml (range 1.28-7.27 nmol/ml) in the control group and the difference was found to be significant (p= <0.0001). Mean serum MDA levels for 1st and 2nd trimester study and control group were 7.60±1.66 vs. 5.00±1.33 nmol/ml and 7.10±2.44 vs. 4.00±1.21 nmol/ml and again the difference was found to be significant (p= <0.0001) between both the participating groups.

Comparison of MDA levels of 1st trimester and 2nd trimester study group was done with the purpose to identify whether OS (oxidative stress) induced risk of miscarriage is higher in early stage of pregnancy and found (Table 7) that serum MDA levels were 7.60±1.66 nmol/ml in 1st trimester miscarriage and 7.10±2.44 nmol/ml in 2nd trimester and difference was found to be non significant (p= 0.4152).

Furthermore, comparison of MDA levels of 1st, 2nd and 3rd miscarriage women with each other was done to check if OS increases with each miscarriage (Table 8). On analysis, serum MDA levels were 7.04±1.53 nmol/ml, 8.53±1.76 nmol/ml, 9.01±2.20 nmol/ml in 1st, 2nd and 3rd miscarriage women respectively and the difference was found to be significant between 1st-2nd miscarriage (p= 0.0198) and non significant between 1st-3rd and 2nd-3rd miscarriage (p= 0.1272, 0.9139).

So as to reveal whether women with declined serum SOD levels are more likely to miscarry and increased SOD levels as an indicator of safe pregnancy, comparison of SOD levels in total, 1st trimester and 2nd trimester study and control group was done. The results showed (Table 9) that mean serum SOD levels in total were 2.36±0.79 U/ml (range 0.70-5.13 U/ml) in the study group and 2.89±1.10 U/ml (range 0.34-5.60 U/ml) in the control group and the difference was found to be significant (p= 0.0016) statistically. Mean serum SOD levels for 1st and 2nd trimester study and control group were 2.18±0.67 vs. 2.72±1.04 U/ml and 2.62±0.89 vs. 3.16±1.15 U/ml

respectively and the difference was found to be significant in 1st trimester and 2nd trimester (p= 0.0085, 0.0431) between both the participating groups.

Likewise, comparison of SOD levels of 1st trimester and 2nd trimester study group was done in order to find whether women with diminished SOD levels are at higher risk for early miscarriage (Table 10). Serum SOD levels were 2.18±0.67 U/ml in 1st trimester miscarriage and 2.62±0.89 U/ml in 2nd trimester miscarriage and the difference was found to be significant (p= 0.0291).

Besides, comparison of SOD levels of 1st, 2nd and 3rd miscarried women with each other was also done in order to determine the depletion of SOD levels with consecutive miscarriages (Table 11). Serum SOD levels were 2.19±0.55 U/ml, 2.04±0.66 U/ml, 1.90±0.63 U/ml in 1st, 2nd and 3rd miscarried women and the difference was found to be non significant among all the three miscarriages (p= 0.1885, 0.6161, 0.1438).

Further, in order to confirm if there is any interrelationship between serum MDA and serum SOD levels in females with spontaneous pregnancy loss, correlation analysis of MDA with SOD in the total, 1st trimester and 2nd trimester study group was carried out and results showed (Table 12) weak negative correlation between serum MDA and serum SOD in total study population(r= -0.3721). As well, in both 1st trimester and 2nd trimester miscarried women, correlation was found to be weak negative (r= -0.4521, r= -0.2209) respectively.

IV. DISCUSSION

The purpose of the current study was to estimate OS in women with miscarriage and to establish MDA as a clinical marker for miscarriage and SOD levels as an indicator for safe pregnancy.

Considerable facts implicate oxidative stress (OS) in the pathophysiology of many complications of human pregnancy, and this topic has now become a major focus of both clinical and basic scientific research. OS arises from imbalance between prooxidant molecules generated from aerobic metabolism and protective antioxidants, influencing the entire reproductive span of the women's life [15]. Although, reactive oxygen species (ROSs) play an important role as second messengers in many intracellular signaling cascades aimed at maintaining the cell in homeostasis with its immediate environment; higher levels can cause indiscriminate damage to biological molecules, leading to loss of function and even cell death [16]. OS is manifested at the maternal-fetal interface from early pregnancy onwards; plays a role in both the normal development of the placenta as well as in the pathophysiology of pregnancy related complications. Placental OS cause widespread destruction of the trophoblastic tissues that is incompatible with ongoing pregnancy and hence spontaneous pregnancy loss [17, 18, 19].

In present study, comparison of malondialdehyde (MDA) and superoxide dismutase (SOD) was done between study and control group in order to elucidate the role of OS in spontaneous abortion and significantly higher circulating levels of MDA (Oxidative Stress Biomarker) were observed in total, 1st trimester and 2nd trimester (p= <0.0001) women with spontaneous pregnancy loss on comparison with their respective controls (Table 6) indicating that systemic oxidative stress, of which lipid peroxidation represents a major manifestation may play an important role in spontaneous abortion and these were consistent with the results of Sumitha Prabhu *et al.*

(2015) and Ghneim and Alshebly, (2016) [20, 21]. Likewise, Abdul-Barry *et al*. (2011) found significantly higher serum MDA levels [8] and lower SOD activity (Ozkaya *et al.*, 2008) in patients with a history of recurrent spontaneous abortion than women with a healthy pregnancy [22] further supporting current findings. As, MDA is a byproduct of lipid peroxidation, thus an acclivity in MDA levels may reflects an overproduction of lipid peroxides and/or impaired antioxidant defense mechanism and increased concentration of lipid peroxides in the villous and decidua of women undergoing early pregnancy loss [23, 24].

When OS develops too early in pregnancy, it can impair placental development and/or enhance syncytiotrophoblastic degeneration, heightened sensitivity of syncytiotrophoblast to OS during the 1st trimester, and could contribute significantly to idiopathic RPL [25] and this was evident in present study also with non significant difference of serum MDA levels in women with miscarriage during 1st and 2nd trimester (p= 0.4152) (Table 7). On the other hand, OS increased with each miscarriage and significant difference was observed in MDA levels between 1st and 2nd miscarriage (p= 0.0198) (Table 8). Although, difference of 1st-3rd miscarriage and 2nd-3rd miscarriage was non significant (p= 0.1272, 0.9139); but role of increased OS with each miscarriage cannot be denied.

Raised antioxidants levels have been documented in healthy pregnancies, while diminished levels were found in women with repeated miscarriages as a consequence of their increased utilization. [26] and this is in agreement with the results of current study; showing significant reduction in serum SOD levels of total (p= 0.0016), 1st trimester (p= 0.0085) and 2nd trimester (p= 0.0431) women with miscarriage on comparison with healthy pregnant controls (Table 9) indicating that OS in terms of decreased SOD activity might be responsible for termination of spontaneous pregnancy loss. Previous studies have also shown that SOD levels were significantly lower in women whose pregnancies ended in miscarriage than in healthy pregnant women [27] and an association of elevated plasma MDA levels along with decreased levels of SOD and other antioxidant enzymes with enhanced lipid peroxidation [28]. Moreover, El-Far *et al.* (2007) found significantly low levels of the antioxidant enzymes GPx, SOD, and catalase in patients with idiopathic RPL, in addition to increased MDA levels, confirming the present findings. [29]

Superoxide dismutase activity might be important for corpus luteum activity, embryonic development and maintenance of early pregnancy. SOD activity has been reported parallel to serum progesterone concentrations in early pregnancy [30]. Progesterone induces decidualization of the endometrium, and there is now some evidence that decidualization induces SOD expression as well [24]. SOD probably works in ovary against the inhibitory actions of peroxide on gonadotropic hormone action, steroidogenesis, and loss of follicular function [31] therefore, the exhaustion of SOD could also be as a result of increased free radical production in terms of raised levels of MDA as pregnancies that went successfully to term were reported to be associated with increased plasma levels of SOD early in the first trimester [27]. Although, the exact mechanism responsible for the spontaneous expulsion is not clearly elucidated but it hypothesized that superoxide radicals stimulate decidual apoptosis, which in turn accelerates the termination of failed pregnancy. Although it has been shown that women with naturally higher levels of antioxidant enzymes are less likely to miscarry [32], the impact of

periconceptional antioxidant supplementation on early pregnancy in the general population remains to be investigated.

Further, serum SOD levels were found to be significantly (p= 0.0291) lower in women with 1st trimester miscarriage than in women with second trimester miscarriage (Table 10); indicative of higher risk in early pregnancy as consistent with findings of Jauniaux *et al.* (2000) that the risk is higher in early gestation because if immature maternal-fetal circulation occurs in early pregnancy [33]; resultant OS cannot be compensated because placental production of protective antioxidants enzymes starts only after 10 weeks of gestation. Additionally, serum SOD levels depleted with each miscarriage; although non significant difference was observed between 1st-2nd, 2nd-3rd and 1st-3rd miscarriage (p= 0.1885, 0.6161, 0.1438) (Table 11), the levels of SOD were definitely lower in 1st trimester and continue to decrease with each subsequent pregnancy, suggesting higher risk of spontaneous pregnancy loss in subsequence pregnancy in women with previous history of loss probably because of depletion of SOD levels to overcome increased OS in each miscarriage.

Correlation analysis of serum MDA with serum SOD showed that there was weak negative correlation of MDA with SOD in total (r= -0.3721), during 1st trimester (r= -0.2209) and 2nd trimester (r= -0.4521) in women with miscarriage (Table 12); reinforcing the observations of [34] Nwadike *et al.* (2017) which showed that increase in the MDA level with concomitant decrease in the antioxidant enzymes, GPx and SOD and the dietary antioxidants vitamins C and E and further supported by El-Far *et al.* (2007), where they measured the MDA and enzymatic antioxidants in patients with idiopathic recurrent pregnancy loss [29]. Therefore, it may be hypothesized enhanced OS in terms of increased concentration of lipid peroxidation markers with diminished SOD either because of decreased production or increased utilization to overcome OS might be responsible for termination of pregnancy. Furthermore, risk of consecutive loss can be reduced by assessing the reason through biochemical analysis of oxidative stress biomarkers and by giving antioxidant therapy during pregnancy especially in early pregnancy.

V. CONCLUSION

So from the present study, it can be hypothesized that enhanced oxidative stress in terms of increased MDA and decreased SOD levels might results in expulsion of the products of conception out of the uterine cavity. The possible reason may be that raised oxidative stress result in placental degeneration with complete loss of syntiotrophoblastic function and detachment of the placenta from the uterine wall and thus miscarriage. Recently, it has been reported that use of antioxidants before conception and during pregnancy significantly improves the chances of live births and outcome in general, but no study has been done so far showing status of oxidative antioxidant ratio in different trimesters and subsequent miscarriages. Hence, future studies with emphasis on OS in women with even first miscarriage are needed to prevent subsequence OS induced spontaneous loss of fetus. Moreover, antioxidant supplementation in women with spontaneous pregnancy losses are required to be recommended to explore the effectiveness of both enzymatic and non enzymatic antioxidants to reduce the risk of OS induced miscarriage.

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Table 1. Comparison of area wise distribution of study and control group

Category	Study group	Percentage	Control group	Percentage	χ^2 value	p value
Rural	42	60%	37	53%	0.7263	0.3941 [NS]
Urban	28	40%	33	47%		

Table 2. Comparison of age at marriage in total, 1st trimester and 2nd trimester study and control group

Parameter	Category	Study group	Range	Control group	Range
Age at marriage (years)	Total	21.44± 3.39	15-33	22.01±3.08	16-31
	1st trimester	21.83± 3.56	15-33	21.97±2.93	17-28
	2nd trimester	20.90± 3.121	16-30	22.07±3.33	16-31

Statistical analysis

Comparison	t value	p value
Total vs. control	1.0370	0.3016 [NS]
1st trimester vs. control	0.1995	0.8424 [NS]
2nd trimester vs. control	1.3840	0.1719 [NS]

Table 3. Comparison of age at 1st pregnancy in total, 1st trimester and 2nd trimester study and control group

Parameter	Category	Study group	Range	Control group	Range
Age at 1st	Total	23.37±3.60	17-34	23.50±2.86	18-32

pregnancy (years)	1st trimester	23.73±3.71	18-34	23.38±2.72	18-29
	2nd trimester	22.86±3.44	17-33	23.66±3.09	18-32

Statistical analysis

Comparison	t value	p value
Total vs. control	0.2326	0.8165 [NS]
1st trimester vs. control	0.4790	0.6334 [NS]
2nd trimester vs. control	0.9242	0.3594 [NS]

Table 4. Comparison of BMI in total, 1st trimester and 2nd trimester study and control group

Parameter	Category	Study group	Range	Control group	Range
BMI (Kg/m^2)	Total	21.33±4.18	12.50-35.41	20.12±3.15	15.07-29.64
	1st trimester	21.17±4.37	12.50-35.41	19.85±2.91	15.07-26.61
	2nd trimester	21.54±3.95	14.44-27.68	20.48±3.46	15.23-29.64

Statistical analysis

Comparison	t value	p value
Total vs. control	1.9260	0.0563 [NS]
1st trimester vs. control	1.6150	0.1110 [NS]
2nd trimester vs. control	1.0900	0.2803 [NS]

Table 5. Comparison of WHR in total, 1st trimester and 2nd trimester study and control group

Parameter	Category	Study group	Range	Control group	Range
WHR (cm)	Total	0.91±0.04	0.80-0.97	0.91±0.04	0.80-0.97
	1st trimester	0.92±0.03	0.85-0.97	0.91±0.03	0.86-0.97
	2nd trimester	0.90±0.04	0.80-0.90	0.91±0.04	0.80-0.96

Statistical analysis

Comparison	t value	p value
Total vs. control	0.3123	0.7553 [NS]
1st trimester vs. control	0.3441	0.7318 [NS]
2nd trimester vs. control	0.7774	0.4403 [NS]

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Table 6. Comparison of MDA levels in total, 1st trimester and 2nd trimester study and control group

Parameter	Category	Study group	Range	Control group	Range
MDA (nmol/ml)	Total	7.34±2.02	3.13-13.59	4.61±1.35	1.28-7.27
	1st trimester	7.60±1.66	4.48-13.59	5.00±1.33	1.62-7.27
	2nd trimester	7.10±2.44	3.13-11.48	4.00±1.21	1.28-6.57

Statistical analysis

Comparison	t value	p value
Total vs. control	9.4130	< 0.0001 [S]
1st trimester vs. control	7.6810	< 0.0001 [S]
2nd trimester vs. control	6.1430	< 0.0001 [S]

Table 7. Comparison of MDA levels of 1st trimester and 2nd trimester study group

Parameter	1st trimester	2nd trimester	t value	p value
MDA (nmol/ml)	7.60±1.66	7.10±2.44	0.8220	0.4152 [NS]

Table 8. Comparison of MDA levels of 1st, 2nd and 3rd miscarriage women with each other

Parameter	1st miscarriage	2nd miscarriage	3rd miscarriage
MDA (nmol/ml)	7.04±1.53	8.53±1.76	9.01±2.20

Statistical analysis

Parameter	Comparison	t value	p value
MDA (nmol/ml)	1st miscarriage vs. 2nd miscarriage	2.4780	0.0198 [S]
	2nd miscarriage vs. 3rd miscarriage	1.6500	0.1272 [NS]
	1st miscarriage vs. 3rd miscarriage	0.1150	0.9139 [NS]

Table 9. Comparison of SOD levels in total, 1st trimester and 2nd trimester study and control group

Parameter	Category	Study group	Range	Control group	Range
SOD (U/ml)	Total	2.36±0.79	0.70-5.13	2.89±1.10	0.34-5.60
	1st trimester	2.18±0.67	0.70-3.40	2.72±1.04	0.34-4.94

	2nd trimester	2.62±0.89	1.13-5.13	3.16±1.15	1.42-5.60

Statistical analysis

Comparison	t value	p value
Total vs. control	3.2230	0.0016 [S]
1st trimester vs. control	2.7170	0.0085 [S]
2nd trimester vs. control	1.9900	0.0431 [S]

Table 10. Comparison of SOD levels of 1st trimester and 2nd trimester study group

Parameter	1st trimester	2nd trimester	t value	p value
SOD (U/ml)	2.18±0.67	2.62±0.89	2.2480	0.0291 [S]

Table 11. Comparison of SOD levels of 1st, 2nd and 3rd miscarriage women with each other

Parameter	1st miscarriage	2nd miscarriage	3rd miscarriage
SOD (U/ml)	2.19±0.55	2.04±0.66	1.90±0.63

Statistical analysis

Parameter	Comparison	t value	p value
SOD (U/ml)	1st miscarriage vs. 2nd miscarriage	1.3490	0.1885 [NS]
	2nd miscarriage vs. 3rd miscarriage	0.5140	0.6161 [NS]
	1st miscarriage vs. 3rd miscarriage	1.6460	0.1438 [NS]

Table 12. Correlation of MDA with SOD in the total, 1st trimester and 2nd trimester study group

Correlation	Category	Correlation coefficient (r)	95% CI	
			Lower Limit	Upper Limit
MDA-SOD	Total	-0.3721	-0.5596	-0.1484
	1st trimester	-0.2209	-0.4982	0.0974
	2nd trimester	-0.4521	-0.7023	0.1021

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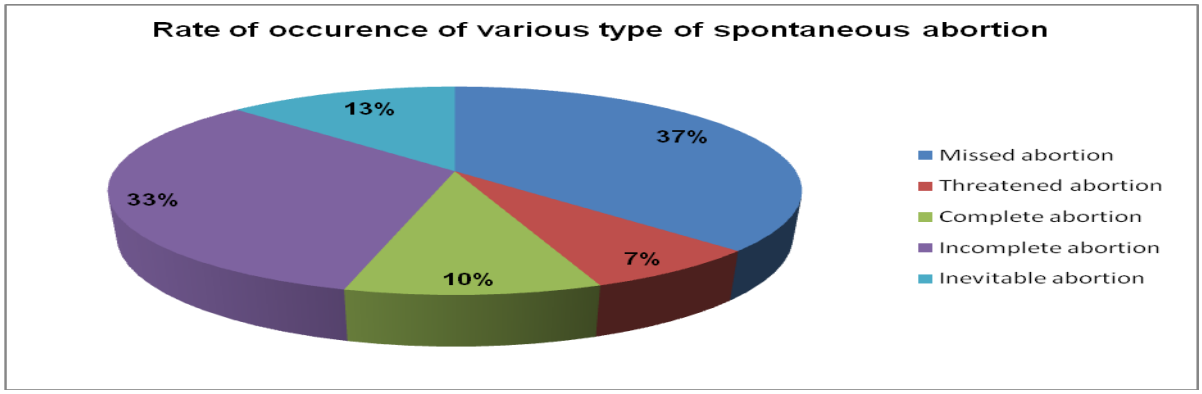

Figure 2. pie chart depicting rate of occurrence of various type of spontaneous abortion

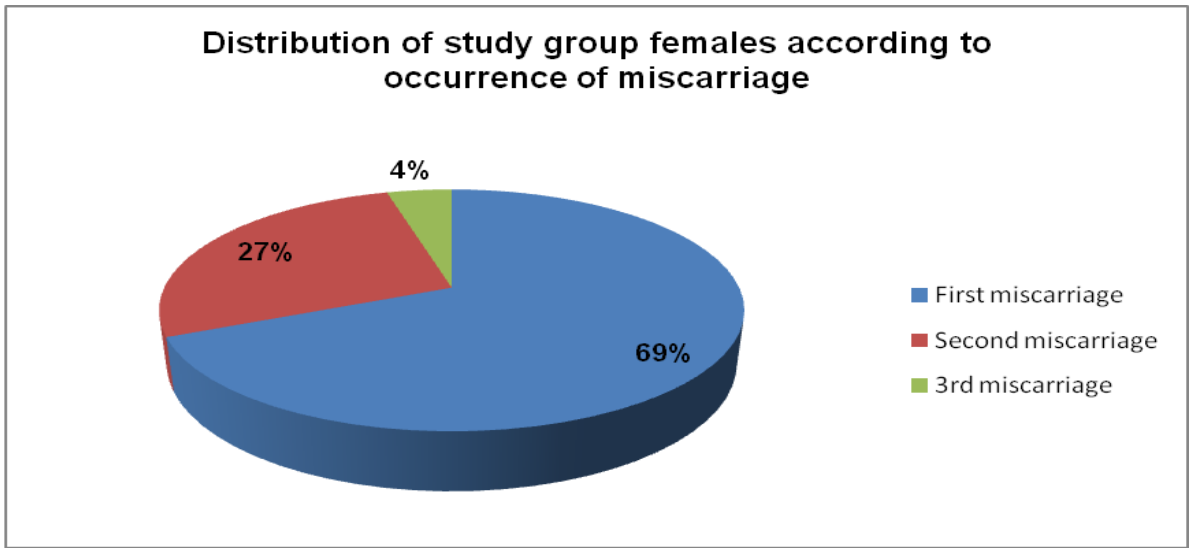

Figure 3. pie chart depicting the distribution of study group females according to occurrence of miscarriage