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Evaluation of Lipid Peroxidation in Cases of Idiopathic Male Infertility

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ABSTRACT

The lipid peroxides and their degradation products are highly toxic to spermatozoa and may play a major role in sperm dysfunction Oxidative stress (OS) is involved in low sperm quality and the etiology of male infertility. The measure of malondialdehyde MDA could be useful diagnostic tool for estimation of oxidative stress. One of the byproducts of lipid peroxidation is malondialdehyde, which has been used as an end product in biochemical assays to monitor the degree of peroxidative damage to spermatozoa. The aim of this study was to determine the level of malondialdehyde in seminal plasma of fertile and infertile men and investigate its relationship with sperm quality. Results showed that the MDA concentration in seminal plasma of unexplained infertile men was significantly higher than fertile men . These finding suggested that oxidative stress was involved in low sperm quality and the etiology of male infertility. The measure of MDA could be useful diagnostic tool for estimation of oxidative stress.

KEYWORDS Seminal plasma, male infertility, sperm quality, malondialdehyde (MDA), lipid peroxidation

Introduction

Defective sperm functions are the most prevalent causes of male infertility and a difficult condition to treat .Many environmental, physiological, and genetic factors have been implicated in the poor sperm functions and infertility . Thus, it is very important to identify the factors/conditions which affect normal sperm functions, along with conventional causes of male infertility such as varicocele, cryptorchidism, infections obstructive lesion cystic fibrosis, trauma a new, yet important cause has been identified, oxidative stress(OS). Lipid peroxidation is a complex process whereby polyunsaturated fattyacids or unsaturated lipids undergo reaction with molecular Oxygen to yield. lipid peroxides. Lipid peroxidation can be defined broadly as "Oxidativedetoriation of PUFA: fatty acid that contain more than two carbon-carbon double bonds"(Halliwell et al., 1984). Lipid peroxidation attacks the fluidity of sperm plasma membrane, with subsequent loss of the ability for oocyte fusion. The propagation of lipid peroxidation through sperm population depends on the antioxidant strategies employed by Spermatozoa and seminalplasma. One of the byproducts of lipid peroxidation is malondialdehyde, which has been used as an end product in biochemical assays to monitor thedegree of peroxidative damage to spermatozoa (Aitken et al., 1989)(2). This assay correlates closely with the degree to which sperm motility and the capacity for oocyte fusion are impaired (Aiiken et al., 1993(1); Siddhu et al.1998)(8) -Mammalian sperm cells present highly specific lipidic composition, high content of fatty acids, plasmalogens and sphinomyelins.

A characteristic feature of most, if not all, biological membranes is an

asymmetrical arrangement of lipids within the bilayer. The lipid composition of plasma membrane of mammalian spermatozoa is markedly different from those of mammalian somatic cells. They have very high levels of phospholipids, sterols, saturated and polyunsaturated fatty acids therefore sperm cells are particularly susceptible to the damage induced by excessiveROS release (Alvarez and Storey, 1995(4). This unusual structure of sperm membrane is responsible for itsflexibility and functional ability of sperm cells. However, spermatozoa lipids arethe main substrates for peroxidation what may provoke functional disorder ofsperm. Low (physiological) level of lipid peroxidation reflects the influence ofreactive oxygen species (ROS) sperm metabolism enhance the ability of human spermatozoa to interact with Zona pelucida. (Aitken et al., 1989). Areas on for lipid peroxidation is oxidative stress. All-lipid components located in the sperm membranes are involved inregulation of sperm maturation, spermatogenesis, capacitation, acrosomereaction and eventually in membrane fusion. Obviously, peroxidation of spermlipids may disturb all the mentioned sperm functions, and in extremecases even completely inhibit spermatogenesis also. The extent to which lipidperoxidation occurs will depend upon the antioxidant strategies available to the spermatozoon. The significance of Lipid peroxidation, and the protective mechanism against ROS has been realized and hence in this investigation, these aspects have been researched to understand the functional alteration ofspermatozoa, which may contribute to poor sperm fertilizing potential

MATERIALS AND METHODS

Detailed investigation on the functional aspects of the spermatozoa in semen of cases unexplained infertility from Ahmedabad and its vicinity was carried out in two groups according to the age range of 20-30(Group II) and 31-40 (Group III).The physical properties of Semen like colour, odour pH showed no significant alteration in all the groups of unexplained infertility as compared to control. The semen quality, judged by ejaculate volume, sperm density, percent motility and morphology of spermatozoa was found to be altered in the cases investigated. The cases under study, were in fact, grouped as Group A and B,based on the sperm density, where Groups IIA and IIIA had counts : comparable to the normospermic range, whereas Group IB and IIIB had Counts in the oligozoospermic range. Semen samples from 46 normozoospermics IIA and III B and 60oligozoospermics,Group IIB and IIIB and 60 were analyzed for physical and biochemical parameters. This data was compared with normal fertile Group 1 Seminal MDA levels were analyzed according to method of Ohkawa et al. (1979(7) is the formation of on a red chromophore that absorbs at 532 nm following the reaction of thiobarbituricacid (TBA) with Malondialdehyde (MDA) and other breakdown products of peroxidised lipids collectively-called as thiobarbituric acid reactive substances (TBARS)

Result

The spermatozoa, unlike other cells, are unique in structure, function and susceptibility to damage by lipid peroxidation (Alvarez et al., 1987). ithasbeen reported that increase in reactive oxygen species (ROS) can cause thedestruction of all cellular structures, including membrane lipids. Hence, in the present study LPO has been used as a marker of oxidative damage to the sperm membrane lipids.

Biochemical evaluation of LPO of each sample from unexplained infertile groups revealed highly insignificant increase ($p<0.001$) in all groups of unexplained infertility investigated (Group II A, II B, III A and III) as compared to the control Groups I and I A. (Tables 1 and 2).

TABLE-1SHOWINGLIPIDPEROXIDATION(LPO)LEVELSINSEMENOFGROUPIIMALES

(CAUSESOFUNEXPLAINEDINFERTILITYOFAGRANGE20-30YEARS)

Group	Parameter
	LPO(nmoles MDA ml/hr)
GroupI Normal n=40	194.2±18.8
GroupII-A Normospermia) n=62	495±17.44**
Group11-B (Moderate Oligozoospermia) n=46	542.5±19.3**

ValuesareMean±S.E.
****p<0.001**

TABLE-1:SHOWING LIPIDPEROXIDATION(LPO) LEVELS IN SEMENOF GROUPMALES(CAUSESOFUNEXPLAINEDINFERTILITYOFAGERANGE3 -40YEARS)

Group	Parameter
	LPO(nmolesMDAml/hr)
GroupI-A Normal n=40	199.8±14.0
GroupIII-A {Normospermia) n=53	297.5±15.3**
Group111-B {Moderate Oligozoospermia) n=59	395.6±18.7**

ValuesareMean±S.E.
****p<0.001**

DISSCUSSION

The spermatozoa, unlike other cells, are unique in structure, function, and susceptibility to damage by lipid peroxidation (Alvarez et al., 1987)(3).It has been reported that increase in reactive oxygen species (ROS) can cause the destruction of all cellular structures, including membrane lipids. Hence, in the present study LPO has been used as a marker of oxidative damage to the sperm membrane lipids. Halliwell (1984) (6) defined lipid peroxidation broadly as oxidative deterioration of PUFA: i.e. fatty acids that contain more than two carbon - / carbon double bonds. LPO attacks the fluidity of sperm plasma membrane, with subsequent loss of the ability for oocyte fusion. The propagation of lipid peroxidation through sperm population depends on the antioxidant strategies employed by spermatozoa and Seminal plasma. One of the byproducts of lipid peroxidation is malondialdehyde, which has been used as an end product in the biochemical assay to monitor the degree of peraoxidative damage to spermatozoa. (Aitken et al., 1989). This assay correlates closely with degree to which sperm motility and the capacity for oocyte fusion are impaired (Sidhu et al., 1998; Aitken et al., 1993)(8,1). Dander et al. (2002)(5) compared EPO with varying sperm count and the poorly swollen sperm (<20%) (HOS test) with highly significant increase in LPO.

The current data too, revealed significant increase in lipid peroxidation, which was correlated with decrease in the sperm functional status as well as the activity of free radical scavenging enzymes viz, SOD and Catalase, Halliwell and Gutteridge (1989)(10)(have also reported an increase in LPO levels and suggested an increase of phospholipase activities during peroxide composition of different Sub organelles and plasma lipids. Reactive oxygen species (ROS) may cause a defect in sperm function through lipid peroxidation. (Sharma and Agarwal, 1996). Therefore, lipid peroxidation and levels of antioxidants have been analyzed in the present study since they are implicated in disturbances of sperm function

Summary and conclusion

Sperm dysfunction caused by reactive oxygen species (ROSs) is one of the major causes of infertility in men, which leads to, lipid peroxidation (LPO) and the formation of stable peroxidation products like Malondialdehyde (MDA) in seminal plasma. MDA is effective factor in reducing fertility MDA is effective factor in reducing fertility In present study, lipid peroxidation (LPO) has been used as a markerof oxidative damage to the sperm membrane lipid. One of the byproducts of LPO is malondialdehyde (MDA), which has been measured as the endproduct in biochemical assay to monitor the degree of peroxidation. Current data revealed significant increase in LPO. Determining the structural andfunctional changes in Sperm membrane lipids during the process of peroxidation is useful in understanding the role of lipid metabolism in spermatozoa. This may help to develop the therapeutic strategies for male infertility. Future research should be directed towards understanding the role of particular components of sperm membrane .

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