Since the discovery of radiation and radioactivity, considerable information has become available concerning the characterization and quantification of radiation and its interaction with matter. Human beings have a substantial risk of exposure to ionizing radiation therapeutically, occupationally or accidently. According to the United Nations Scientific Committee on the Effects of Atomic Radiation (UNSCEAR, 1982) report, largest contribution to ionizing radiation exposure to men is through its diagnostic and therapeutic uses. Generally, radiation health workers are employees of hospitals and private clinics where radiation is used for diagnostics and therapeutic purpose to humans. There are approximately 7 million radiation health workers worldwide including India (UNSCEAR, 2008). Thus, hospital workers are constantly exposed to very low doses of ionizing radiation throughout their reproductive age which is one of the major concerns.

The effects of radiation on reproduction are well established as early as in 1920’s. The reproductive organs have the highest sensitivity to radiation which is represented by the highest weighting factor of 0.20 (ICRP, 1991; NCRP, 1993), which may be because of presence of highly proliferating tissue compartments in the reproductive organs especially, testes & ovary. In other words, a certain amount of radiation exposure to reproductive organs produces much more serious health effects than the same amount of exposure to other organs. Decades of animal and human research have convincingly documented that a variety of risk factors including exposure of a male to ionizing radiation can cause genomic instability (Balakrishnan and Rao, 1999; Cigarran et al., 2001; Tawn and Whitehouse, 2003), alter cellular and humoral immunity (Godekmerdan et al., 2004; Serhatlioglu et al., 2004), impair fertility (Wyrobek et al., 1997; Ahamadi
and Ng 1999) and affect reproductive outcome (Mole et al., 1966; Baker 1971; Saharan et al., 1977; Adiga et al., 2007a,b; Adiga et al., 2010).

In human, extensive laboratory data has shown the detrimental effect of ionizing radiation exposure on spermatogenesis (Fischbein et al., 1997). A report by International Atomic Energy Agency suggests that maturing spermatogonia are about 70 times more sensitive to radiation damage than the stem cell (IAEA, 2004). Therefore, long term exposure to radiation affects the sperm cells which can lead to abnormal spermatogenesis. Hence, chronic occupational radiation exposures to male health workers are one of the major concerns.

Mechanism of action of radiation and their cellular effects are well established. Exposure to ionizing radiation induces radiolysis of H₂O into H⁺ and OH⁻ radicals. These radicals are chemically very reactive, and produce a series of highly reactive combinations such as superoxide (HO₂) and peroxide (H₂O₂). These chemicals further induce oxidative damage to molecules within the cell. Ionizing radiation can induce DNA damage directly by acting on nucleus or indirectly by inducing free radicals in the cell which will ultimately act on DNA. The interaction between the ionizing radiation and cellular DNA produces DNA damage including single strand breaks (SSBs), double strand breaks (DSBs), damage to nucleotide bases, and cross links between DNA-DNA and DNA-protein.

Fertilization, embryo quality and health of the offspring can be affected by defect in the sperm chromatin (Haines et al., 2002). Previous studies from our laboratory have
observed that exposure of testicular regions of male mice resulted in sperm DNA damage in a dose-dependent manner and analysis clearly demonstrated a significant increase in the DNA damage even in spermatozoa exposed to the lowest dose of radiation (Adiga et al., 2007, 2010; Kumar et al., 2013). Moreover, transgenerational changes in genomic instability in the pre-implantation embryos derived from irradiated sperm was observed, suggesting that radiation induced sperm DNA damage can cause genomic instability in pre-implantation embryos (Adiga et al., 2010). In addition, a dose dependent decline in number of litters and high incidence of post-natal death was observed in the progenies derived from irradiated spermatozoa in mice (Kumar et al., 2013). However, till now there is no study to find out the effect of occupational radiation exposure on the sperm functional characteristics and DNA integrity/fragmentation in the radiation health workers who are exposed to low levels of radiation.

Epigenetic modification is a collection of mechanism and phenomena that define the phenotype of a cell without affecting the genotype and it is very important for the exchange of histone to protamine in the spermatids, differential gene expression during fertilization and reprogramming of epigenetic information during embryogenesis (Bird, 2002; Feinberg, 2007; Umlauf et al., 2008). There are few major concerns about the transmission of epigenetic abnormalities, such as Angelman and Beckwith-Wiedmann syndromes through assisted medical conception (Gosden et al., 2003; Debaun et al., 2003; Marques et al., 2004). In addition, the environment around the living place and/or workplace has considerable effects on the epigenetics of adult individuals (Aquilera et
al., 2010). Till now, there is no report on the effect of occupational radiation exposure on the global DNA methylation of the spermatozoa in radiation health workers.

Semen is a mixture of spermatozoa suspended in secretions from testes and epididymis which, at the time of ejaculation, are combined with secretions from prostate, seminal vesicles and bulbourethral gland, collectively known as seminal plasma. It plays an important role in various sperm function like maturation, capacitation and fertilization. It is well endowed with an array of enzymatic and non-enzymatic enzymes such as catalase, superoxide dismutase (SOD), glutathione peroxidase/reductase pair (Alvarez et al., 1987) and ascorbic acid, uric acid, thiols (Lewis et al., 1997) respectively, which protects the spermatozoa from reactive oxygen species (ROS) (Kankofer et al., 2005). As long as spermatozoa are suspended in seminal plasma, they are protected from oxidative damage by lipid peroxidation. Previous studies have shown the enhanced antioxidant concentration in the blood plasma to chronic exposure in health workers (Durović et al., 2008; Russo et al., 2012) but there is no data available to find out the effect of occupational radiation exposure in the seminal plasma that links the effects of occupational radiation on antioxidant level in the seminal plasma and their association with sperm DNA damage in the health workers.

The proliferation, differentiation and survival of the germ cells depend upon on gonadotropin-dependent mechanisms (McLachlan et al., 2002a). Mitotic and meiotic divisions require master regulators such as follicle stimulating hormone (FSH), testosterone and leutinizing hormone (LH) from the hypothalamo-pituitary-testicular (HPT) axis (Ruwanpura et al., 2010). FSH supports the spermatogonial development
whereas, testosterone partly supports spermatocyte maturation (McLachlan et al., 2002a; Ruwanpura et al., 2008a,b) and spermiation. However, both testosterone and FSH is required for the release of spermatids from Sertoli cell (McLachlan et al., 2002a). LH mainly acts on the Leydig cells which synthesize the testosterone (Wahlstrom et al., 1983). Reproductive hormones are considered as one of the predictor of semen quality in epidemiologic studies. Semen quality is associated with changes in reproductive hormones in men (Jensen et al., 1997; Mahmoud et al., 1998; Meekar et al., 2007) and therefore, serves as a substitute for the assessment of male reproductive function (Uhler et al., 2003). The potential impact of chronic occupational exposure to various hazards agents can lead to damage to sperm chromatin, decrease in semen quality and alteration in reproductive hormones (Yucra et al., 2006; Miranda et al., 2013) which essentially reflects primary and secondary effect on occupational hazards on the testes and the HPT axis. Most of this physical and chemical occupational hazards acts on the male reproductive system by disrupting the HPT axis. However, so far there is no report on the effect of occupational radiation exposure on the reproductive hormone level of the male health workers.

As per PUBMED literature survey, we are not aware of any studies done among Indian (Non-Caucasians) radiation health workers for the evaluation of sperm characteristics and reproductive toxicity. Therefore, this study was planned to evaluate sperm abnormalities, serum reproductive hormones, global methylation pattern and plasma antioxidants in the health workers occupationally exposed to radiation at their workplace and then find out its association with the level of radiation exposure level as measured by the radiation
dosimetry. Primary objective of this study was to understand the influence of radiation exposure on the sperm functional, genetic, epigenetic and endocrine hormones in the subjects who are handling radiation sources at their workplace. The objectives of this study were as follows:

1. To study the prevalence of semen abnormalities, sperm functional characteristics in the subjects who are handling radiation sources at their workplace.
2. To analyze the incidence of DNA damage/ chromatin abnormality/ aneuploidy in spermatozoa and peripheral blood lymphocyte in radiation health workers.
3. To evaluate the general methylation status in spermatozoa of radiation health workers.
4. To quantify the level of antioxidants and reactive oxygen species (ROS) in the seminal plasma of radiation health workers.
5. To assess the possible influence of occupational radiation exposure on serum reproductive hormones such as FSH, LH, and testosterone.