SUMMARY AND CONCLUSION
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Lead and some of its chemical compounds are virtually ubiquitous in the environment. Lead can now be found in air, drinking water, as well as in soil and various food items. The possibility of lead exposure in humans is therefore of great significance from the health point of view. Lead does not spare any organ in the body and is not known to serve any necessary biological function (2). The presence of blood lead has always been considered in a sign of environmental pollution, whether it originates from general or occupational environment. Lead poisoning is one of the major environmental diseases in the world. Its impact on the Indian population is not fully understood. The population of developing nations like India is particularly more susceptible to lead poisoning, because of the problem of malnutrition, iron deficiency anaemia, because more lead is absorbed in this condition and therefore greater toxic effects at the same level of lead exposure. Adults at highest risk are those who are exposed to lead at work such as battery manufacture workers, painters and silver jewellery workers, smelters, printers and plumbers. The clinical diagnosis of lead poisoning in the adults is often complicated by the lack of any clear symptoms and signs.

Lead is highly resistant to corrosion, more pliable, high density, low elastic, high thermal expansion, low melting point, ability to form carbon metal compounds, hold pigment well, easy workability, very easily recycled, excellent antifriction metal and inexpensive. Due to these properties it is used for various purposes, such as in acid batteries, pigments, cable sheathing, petrol additives, solder applied to water distribution pipes, and to seams of cans used to store foods, in paper industries ceramic glazes, ship construction, printing press silver jewellery industries, cosmetics & folk medicines casting toys and ornaments etc. (1, 2).

Air borne lead can be deposited on soil and water thus reaching human through the food chain and in drinking water. Atmospheric lead is also a major source of lead in household dust. Lead contaminated dust is a major source of health risk to children and adult in most domestic and occupational environment. Levels of lead found in air, food, water and soil/dust varies widely throughout the world and depend upon the degree of industrial development, urbanization and lifestyle factors (1,2).

Lead absorbed by the GIT comes from the intake of lead in food, beverages, and soil/dust in case of older children and adults and in occupational exposure population mostly atmospheric air. Dietary factors, nutritional status, chemical form of the metal and patterns of food intake affect adsorption. Absorption of lead is more in fasting condition than in feeding.
condition. Nutritional inadequacies can also affect toxic response to lead. Increased calcitriol concentration increases GIT absorption of lead. Fewer intakes of Ca, Po₄, Fe, Se and Zn increase the absorption of lead (1).

Lead is not distributed homogeneously throughout the body. It is rapidly taken up in blood and soft tissues (half life 28-36 days) followed by a slower redistribution to bone (half life 27 years). In bone more lead is accumulated and may serve as endogenous source of lead. Bone lead is readily mobilized to blood and the effect is most apparent in people with history of occupational exposure. Dietary and airborne lead which is not absorbed in the GIT excreted in the faces. Blood lead is not retained in the body it is excreted in urine & faces (1).

In human, lead can result in a wide range of biochemical effects depending upon the level and duration of exposure. It affects many biochemical processes mainly on haem synthesis, haematopoiesis, nervous system, renal system, cardiovascular system and reproductive system.

The PbB and PbU level were significantly increased in the study groups as compared to control subjects. The PbB levels depend on the relationship between intake, storage, and excretion. Blood lead level is primarily an indication of acute (current) exposure but is also influenced by previous storage. Values more than 40 µg/dl are generally considered abnormal in adults. The symptoms of lead poisoning are associated with levels more than 80 µg/dl, while mild symptoms may occur at 50 µg/dl in children and adults. The acute symptoms of lead poisoning such as abdominal colic, constipation, anorexia, vomiting, headaches, anxiety, depression, irritability, excessive sweating, muscle and joint pain. These symptoms were observed in most of battery manufacture workers, and silver jewellery workers.

The PbB level is good and most sensitive biomarker for screening the lead exposure population and its adverse effects, but the half-life of lead in blood is short (28-36 days). Therefore, the estimation of PbB level generally reflects only recent exposure. Also a single PbB measurement cannot differentiate of low-level chronic exposure from a short high-level exposure, because cycling of lead between blood, bone soft tissues.

The PbU excretion has also been used as an index of exposure, since PbB values change more rapidly than PbU excretion. However, excretion values depend on 24-hour urine specimens, with the usual difficulty in complete collection. Also so many factors affects on the PbU excretion such as renal function, fluid intake and the specific gravity of the urine.
Therefore, PbU estimation appears to be limited use for general screening. But PbU levels after administration of the chelating agent like CaNa$_2$-EDTA is considered an excellent measure of the potentially toxic fraction of the total body burden of lead (1). The PbU excretion half-life is approximately 45 days (329), and excretion rates increase during chronic exposure to inorganic lead (327).

The PbU level was significantly increased in battery manufacture workers and spray painters as compared to normal control, but no significant difference was observed in case of silver jewellery workers, though PbB level was high. It may be due to more excretion of lead through sweating, because silver refining unit workers were exposed to high temperature. Whole work place was very hot, due to inadequate ventilation and no chimney was fixed at silver refining unit. Poor positive correlation was found between PbB and PbU in battery manufacture and silver jewellery workers and good positive correlation in case of spray painters ($r = 0.78$). It may be due to the wide variation of PbB level in these two study groups.

Increased PbB values in study groups as compared to the control group indicate the rate of lead absorption is definitely more in all the 3 study groups. However, the PbB level is high in battery manufacture and silver jewellery workers. Battery recycling and manufacturing is an informal work. Lead is used for making grids, bearings and solder purposes, manufacturing processes are usually manual and involve the release of lead particles and lead oxides that may cause severe poisoning and environmental pollution. Battery recycling is an important source of exposure to inorganic lead vapors, particles and debris, poor hygiene and inappropriate protection increase the risk of exposure. Exposed workers also pose a source of exposure to their family members. In silver jewellery industry, lead is mainly used for making design on silver ring, for this study mainly workers engaged in making such type of rings and silver refining units workers were included. The work atmosphere was filled with lead and lead oxide fumes lasting for 4-6 h/day/week. The vapors also condensed on workers skin, hair, contaminating their clothing, food, and drinking water. Also this type of work is predominantly carried out at home or in non-regulated shops. The lead fumes and dust generated in silver refining units and silver rings making workshops pose an exceptional health hazard to children and adults living near these operations.

In spray painters the PbB level is increased as compared to normal control subjects but not increased more like other two study groups.
It may be due to less use of lead in paints or ban on addition of lead in paints. Nowadays lead-free or low lead content paints are available in market. Lead is used in various paints because of its anticorrosive properties. In India 10 % of total lead metal utilised is used in the manufacture of paints. Wherever such paints are used there will be the potential for human lead exposure.

The non-activated δ-ALAD was significantly decreased (P < 0.05) in battery manufacture and silver jewellery workers as compared to the control subjects. However, activated δ-ALAD mean values were not statistical significant, but the ratio of activated/non-activated δ-ALAD was significantly increased (P<0.001) in all the three study groups as compared to the controls. The erythrocyte δ-ALAD is activated by Zn, acetate, because this is a zinc dependent metalloenzyme and zinc partly protects the enzyme against the adverse effects of lead in Vitro (330) and possibly also in Vivo (331).

Lead in the blood is said to be rapidly incorporated into RBCs and probably affects δ-ALAD by directly inactivating the sulphhydryl groups necessary for its activity (333). In several experimental lead poisoned animal study were also reported that the activity of δ-ALAD were impaired due to interference of sulphhydryl groups of this enzyme in the brain, liver, Kidney and bone marrow, and addition of glutathione increased the activity by 30-40 % (333-335). The endogenous GSH concentration in erythrocytes is shown to decrease significantly in workers intoxicated by lead (337). Several authors hypothesized that the GSH in erythrocyte or free-SH groups in liver cell plays a crucial role in the regulating of δ-ALAD activity (104).

The non-activated δ-ALAD activity alone is good marker enzyme for lead toxicity, In present study this enzyme is activated by zinc acetate and activated as well as non activated δ-ALAD activities were measured and the ratio of activated /non-activated of δ-ALAD was calculated. Since several studies reported that the erythrocyte δ-ALAD activity is also increased in anaemia and sickle cell disease (338, 339). In present study, it was found that the ratio of activated /non-activated δ-ALAD was significantly increased (P<0.001) in all the three study groups as compared to the controls. This appeared to to be a good marker of lead toxicity as increased ratio is observed in high PbB level. Therefore, it is concluded that the δ-ALAD activity decreases or inhibited by the lead in all the 3 study groups as compared to the controls. This observation is also supported by the good negative correlation coefficient.
values between PbB and non-activated δ-ALAD activity in silver jewellery workers \((r = -0.5126)\) and battery manufacture workers \((r = -0.3906)\).

The non-activated δ-ALAD activity was not significantly decreased in case of spray painters as compared to control. And also we found very weak negative correlation between PbB and non-activated δ-ALAD in spray painters group. This may be due to less PbB level in spray painters as compared to other two study groups. The δ-ALAD activity is reduced to 50 %, when PbB values are in 30-50 μg/dl range, reported by several studies. Therefore, the estimation of δ-ALAD activity in erythrocyte is very good, sensitive, most reliable, marker enzyme for screening in occupational lead exposure.

The urinary δ-ALA and PBG concentrations were measured in study groups and the controls. Significantly increased excretion of δ-ALA and PBG were found in urine in all the 3 study groups as compared to the controls. Also the good positive correlation coefficient was observed between PbB and U-δ-ALA in all the 3 study groups. However, there was no significant correlation was found between PbB level and U- PBG excretion in all the 3 study groups.

In several studies, it was reported that the lead inhibits the δ-ALAD enzyme, resulting the increased level of δ-ALA in the blood, which will then increased excretion of δ-ALA in urine \((111 \text{ to } 113)\) and the excretion of δ-ALA in urine will be more at PbB level of around 35 μg/dl \((112)\). Similar results also reported in lead poisoned rabbits \((111)\). Therefore of urinary δ-ALA and PBG are the good indicators of body lead burden, and also it is indirectly useful to known the inhibition of δ-ALAD enzyme activity and PbB level in lead exposure population.

The RBC zinc protoporphyrin concentration was significantly increased only in battery manufacture workers as compared to control group. But no significant correlation coefficient was found between PbB and RBC-ZPP in this group. This may indicate a long, intense lead exposure in battery manufacture workers. This finding is consistent with other reports \((340-345)\). Lead inhibits ferrochelatase enzyme, which incorporates iron into protoporphyrin IX to form heme. Impaired protoporphyrin IX conversion leads to increased erythrocyte protoporphyrin levels, which binds with zinc to form ZPP, measured by hematofluorometer instrument. The RBC-ZPP is not good marker for screening the lead exposure population because, it is increases, in chronic iron deficiency haemolytic anaemia, erythropoietic protoporphyria, chronic febril illness, increase serum bilirubin level \((1,2)\). Also

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it is not sensitive enough for lead screening in children because it is elevated only at high PbB levels (40-60 µg/dl).

Haematological parameters such as Hb, PCV, MCV, MCH, MCHC were significantly decreased in battery manufacture workers as compared to normal control. Whereas these haematological parameters are not significantly changed in other two study groups except RBC count, which is decreased in silver jewellery workers. The total WBC count was significantly increased in battery manufacture and silver jewellery workers.

Several studies reported that the anaemia is common in high lead exposure population (117, 118). It is due to impairment in the synthesis of heme, impaired maturation of RBCs and Hb deficiency. Lead affects the hemopoietic system and decreases the Hb synthesis. It might be due decreased haem and globin synthesis or erythrocyte formation and function. The rate of erythrocyte formation is regulated by erythropoietin hormone and the serum level of this hormone is decreased by the lead (1).

Lead poisoning may be associated with a hypochromic microcytic anaemia, although more commonly, there is a normochromic, normocytic anaemia. Lead anaemia has many of the features of a typical sideroblastic anaemia for example impaired maturation, and defective haemoglobinization of RBCs, raised serum iron, hypochromia and erythroblasts containing iron staining inclusion bodies (122).

In case of battery manufacture workers, a good negative correlation was observed between PbB and haematological parameters such as Hb (r = -0.59), PCV (r = -0.61), MCV (r = -0.47), MCH (r = -0.46), and in spray painters group Hb (r = -0.60), MCHC (r = -0.46), while in silver jewellery workers very weak negative correlation were observed between PbB and haematological parameters. These results indicate that the anaemia is common in battery manufacture worker at high PbB level. In spray painters, it may be due solvents used in spray painting which alters the haematological parameters. Therefore, the estimation of haematological parameters are useful for screening the occupational lead exposure in the first 2 years of the job of workers.

The biochemical parameters of liver function i.e., SGOT and SGPT levels were significantly increased only in spray painters and there was no significant difference observed in other two study groups as compared to the controls.
The mean values of SGOT and SGPT are at higher side of normal range and high variation of these values also observed in spray painters. Therefore, the increased transaminase activity in spray painter is not due to the lead exposure per se, it may be due to the solvents (xylene and toluene) used for spray painting that are known as hepatotoxic chemicals. Therefore, the detailed studies are required to find out the exact cause of increased transaminase activity by lead.

Decreased serum total proteins level was observed in all the 3 study groups. The mean values of serum albumin was decreased only in silver jewellery workers and spray painters, serum globulin concentration were significantly decreased only in battery manufacture workers, and the A/G ratio is significantly increased in battery manufacture and silver jewellery workers. The A/G ratio was not different in spray painters as compared to the control subjects.

This observation indicates that the decreased protein synthesis, mainly albumin and globulin at high PbB levels in battery manufacture and silver jewellery workers. The A/G ratio is significantly increased in battery manufacture and silver jewellery workers Whereas A/G ratio was not significantly different in spray painters as compared to the control subjects. The A/G ratio is increased in these two study groups may be due to more decreased globulin synthesis than the albumin at high PbB levels. The A/G ratio did not alter in spray painters may be due to low PbB level. This observation has supported previous report in experimental animals and lead exposure. The globin synthesis is inhibited by lead in rat bone marrow cell at 1 μmol/L concentrations (111), therefore the measurement of total proteins also an important parameter to detect the impairments of liver function in high lead exposure workers.

The serum bilirubin level was slightly increased only in battery manufacture workers, while there was no significant difference observed in other two study groups as compared to the control subjects.

Increased serum bilirubin level in lead poisoning cases reported in several studies (358 to 363). The high concentrations of lead have been shown to produce morphological changes and destruction of red cells, when administered in vitro or vivo (364). Therefore, the evidence of various reports and present results suggest that the rate of haemolysis is more at high PbB level. Hence, the serum bilirubin level is increased only in battery manufacture workers as compared to control group.
The biochemical parameters of kidney function i.e., the blood urea levels were significantly increased only in battery manufacture workers, decreased in silver jewellery workers, and no significant difference found in spray painters as compared to control. The serum uric acid level was decreased only in silver jewellery workers and no significant difference observed in other two study groups as compared to control. The serum creatinine level was not altered in all the 3 study groups as compared to control group.

Lead is known to cause proximal renal tubular damage, characterized by generalized aminoaciduria, hypophosphataemia, with relative hyperphosphaturia and glycosuria accompanied by nuclear inclusion bodies, mitochondrial changes and cytomegaly of the proximal tubular epithelial cells. Tubular effects are noted after relatively short-term exposures and are generally reversible, whereas sclerotic changes and interstitial fibrosis resulting in decreased kidney function and possible renal failure require chronic exposure to high lead levels. Increased risk of nephropathy was noted in workers with a PbB level of over 60 µg/dl. (1). Therefore, measurements of kidney function test donot have importance at low PbB level. We found the mean value of blood urea at higher side of normal range in battery manufacture workers, may be due to high PbB level. In other two study groups no significant change in renal functions parameters as compared to the controls was found at low PbB level.

In all the three study groups, serum lipid peroxidation levels were significantly increased and antioxidant enzyme activity such as SOD, Catalase, Ceruloplasmin were significantly decreased, except ceruloplasmin in spray painters, which was not changed as compared to control. While other antioxidant parameters such as uric acid and albumin concentration were significantly decreased in silvers jewellery workers and spray painters as compared to control subjects. These observations are consistent with earlier reports in the literature.

In several studies reported that the Lead causes oxidative stress. The mechanism of Pb toxicity could be expressed through by inducing the generation of reactive oxygen species, reducing the antioxidant defense system of cells via depleting glutathione, inhibiting sulfhydryl-dependent enzymes, interfering with some essential metals needed for antioxidant enzyme activities, and/or increasing susceptibility of cells to oxidative attack by altering the membrane integrity and fatty acid composition (14), Lead induced disruption of the Pro-oxidant /antioxidant balance in lead burdened tissue could contribute to tissue injury via
oxidative damage to critical bio-molecules. Significant accumulation of MDA, a by-product of lipid peroxidation for both in vitro & vivo system, has been observed (144).

Very good negative correlation coefficients were found between lipid peroxidation and antioxidants parameters such as SOD, and catalase only in battery manufacture and silver jewellery workers. This observation suggests that at high PbB level the rate of generation of free radicals is more and at same time antioxidant capacity is depleted. While no significant correlation was found between LP and other antioxidants parameters i.e. ceruloplasmin, bilirubin, albumin and uric acid in all the three study groups. It indicates that the primary antioxidant enzymes is mainly utilised against oxidative stress.

From past reports and the present results, it is tempting to speculate that the increased lipid peroxidation and impaired various antioxidant parameters in lead exposure workers might be due lead toxicity. It is not clear whether these alterations are the cause of the oxidative damage or lead effect on the antioxidant defense system of cells because some of the components of the antioxidant defense system that occur first by lead and that might cause an impairment in pro-oxidant/antioxidant balance of cells resulting in oxidative damage. Therefore the administration of antioxidant vitamins like ascorbic acid, α-tocopherol, β-carotenes and GSH will be beneficial to lead exposed workers.

In conclusion, it may be said that in some industries where lead is involved e.g. Battery manufacture, Silver jewellery and Spray painting and like industries despite modern technical advancement considerable lead hazards still exist. This research work is able to demonstrate that complete hemogram, urinary δ-ALA, PBG and erythrocyte δ-ALAD estimations are most valuable in screening the occupational lead exposure.

There was no evidence of severe disturbance of liver functions in the occupational lead exposure such as in battery manufacture, silver jewellery work and spray-painting.

Similarly there was lack of evidence indicating severe impairment in kidney functions in the occupational lead exposure such as in battery manufacture, silver jewellery work and spray painting.

There is imbalance of pro-oxidants / antioxidants in occupational lead exposure, indicating oxidative stress induced by increasing lead pollution. Such workers require additional dietary supplements of antioxidants along with other protective measures.
Medical examination and investigations should include specific tests like blood lead level, urinary δ-ALA, PBG and erythrocyte δ-ALAD estimations along with routine tests. These check ups to be done not only at pre-employment stage but also at regular intervals during their service to identify worker with potential lead toxicity. In addition, environmental lead level should be kept below permissible level.