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# A GENERAL EVALUATION OF HEAVY METALS CONCENTRATION IN THE LEACHATES OF PLASTIC BIOMEDICAL DEVICES

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***ABSTRACT***
*The main objectives of study were to monitor the heavy metals concentration in the leachates of four plastic biomedical devices of 21 brands including IV sets, seven brands, DNS bottles, five brands, RL bottles, five brands and Ryle's tubes four brands used in the present study. Plastic biomedical devices were washed thoroughly with sterilized double distilled water prior to leaching. Aseptic dried plastic biomedical devices were cut into small pieces of 1 $cm^2$ and immersed in 100 ml of either of simulating solvent viz. double distilled water, ethanol (8%), acetic Acid (3%), sodium chloride (0.9%) and sodium carbonate (5%) at 25±2°C for 24 hours (ambient conditions) and 60±2°C for 2 hours (elevated conditions). Parallel sets having simulating solvents only were also run under identical conditions and were served as basal control. The leachates will be taken in flask and digested with concentrated Nitric acid and the volume of digested samples will be made upto 10 ml using 1% Nitric acid. The digested samples were analyzed for metal content, (Cd, Cr, Cu, Fe, Mn, Ni, Pb and Zn) using a Perkin- Elmer atomic absorption spectrophotometer. At 60±2°C for 2h, the highest mean concentration of metals (cadmium, 0.057±0.002; chromium, 0.122±0.002; copper, 0.138±0.004; Iron, 7.476±0.003; nickel 0.313±0.02; lead 0.220±0.002 and zinc, 5.954±0.009 ppm. At 25±2°C for 24h, the highest mean concentration of metals (cadmium, 0.051±0.001; chromium, 0.121±0.009; copper, 0.131±0.002; iron, 2.41±0.001; nickel 0.31±0.004; lead 0.20±0.001; zinc, 1.91±0.002 ppm.*

***Keywords****: Atomic Absorption Spectrometry, biomedical devices, heavy metals, leachates*

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## INTRODUCTION

Plastics are polymeric materials that have remarkably transformed and alleviated our daily lives by replacing many expensive manufacturing raw materials. Their economical and resilient use in automobiles, aerospaces, electronic gadgets, furnitures, kitchen wares, packaging, agricultural and construction materials, telecommunication, health and medical products and many other household and industrial products has increased over the years (Mc Roberts TS 1973). Plastics have many unique properties in terms of their manufacturability and production possibilities, and these are being increasingly utilized in the production of medical devices and medical packaging (Lewis R 1999). Environmental factors such as light, heat, moisture, chemical and biological processes may bring about physical and chemical changes in the polymers causing bond breaking and structural deformations (Maes D 1986, Donnell JH). Such polymer degradation causes cracking, erosion, discoloration and delamination etc (Viletti MA 2002).

The finished plastics are generally considered to be safe provided they are manufactured at standard conditions using permitted chemicals recommended by national and international regulatory agencies and used properly (BIS 1981, 1986, ISO 1997, USP 1995, BP 1998). In the present scenario, we find the plastic usage pattern such as in various sectors like 33% being used for packaging , 20% in building construction, 10% in electrical and electronics, 7% in automobiles, 5% in agriculture and 25% in the other sectors such as medical. Plastic use is dominated by single use or short term use, and at the same time most plastics are extremely persistent in the environment. Polymer scientists, working closely with those in the device and medical "fields, have made tremendous advances over the past 30 years. Demand is expected to increase until 2015, growing at a CAGR of 9.8% from 2009-2015. Existing polymers will continue to be used in medical applications, while technological advancements will help to improve their properties. Other forms of plastics will be developed for medical use, thereby leading to an increase in demand for medical applications. In hospital, plastic medical devices are being used for storage and transfusion of life saving fluids, syringes, blood bags, biomedical implants, tubing and heart valves for the cardiac patients. North America leads the demand for medical polymers with a share of 42.2%, Europe 30.1% and Asia 22.8%. Polyvinyl Chloride based medical polymers have the largest share when it comes to medical applications. Properties such as softness, high transparency, facility of sterilization and strength make PVC the first choice for medical polymer manufacturers. PVC based medical polymers constitute 40%, Polypropylene 20% followed by Polyethylene with a share of 15%. Other medical polymers had a share of 25%. Prominent other plastics included Polycarbonate, Acrylonitrile, Styrene Butadiene, Biopolymers, Nylon and Polystyrene. Polymers used in medical packaging applications need to be transparent or translucent, and in addition, to have antibacterial properties. Many inorganic chemical additives can be added to finished plastic biomedical devices in order to get desired physical, chemical, or mechanical properties. These include stabilizers, fillers, plasticizers, pigments, antioxidants and flame retardants etc. (Deanin RD 1975, Arnold LK 1968). The common use of various inorganic elements such as Al, As, Ba, Br, Ca, Cd, Co, Cr, Cu, Fe, Hg, Ni, Pb, Sb, Sn, Se, Ti and Zn etc. as these additives in plastics is also of concern from human exposure and pollution point of view as many of these elements are hazardous for human health. These additives are not chemically bound to the matrix of the polymeric materials and leach out under the influence of physicochemical factors such as sun light, temperature, type of solvents and pH of the stored commodity (Figge K 1977, Srivastava SP 1984, Parmar D 1985, Khaliqui MA 1992, Alam MS 1990, Junaid M 1998, Sharma VP 1998, Jenke D 2011, Jenke D 2010, Jenke DR 2007, Jenke D 2012). Release of hazardous substances from plastic products to air, extraction fluids, water, food, food simulants, saliva and sweat have been shown by chemical analysis such as lead, tin and cadmium etc.(Al-Malack, 2001). Compounds that include organics, metals and volatile sulphur containing compounds may leach out from the plastic IV infusion containers into the contained solution (Gallelli JF 1993, Jenke DR 2002, Jenke DR 2005).

Zinc based organic compounds are often used to initiate polymerization and wide ranging trace levels of zinc are found in IV bag diluents (Desai N 2007). There are numerous reports on the leachable, such as metals, DEHP, cyclohexanone and other organic and acidic compounds found in the solution of PVC bags (Chawla AS 1991, Arbin A 1986, Cheung A P 1998, Demore B 2002, Pearson SD 1993, Ulsaker GA 1977). Heavy metals, such as Cd, Cr, Cu, Fe, Mn, Ni, Pb and Zn leaching in the simulating solvent are known to be present in the finished plastic products (Srivastava SP 1984). They affect the normal physiological activities of the cells, as the permissible limits. Chronic low level toxicity of lead and cadmium came into view in recent years as a problem of our civilization.

General health effects to be considered associated with exposure to these metals comprise systemic, immunological, neurological, reproductive (Benoff S 2000), developmental, genotoxic and carcinogenic, (Hartwig A 2010) and even death (ATSDR 2008). The metals can induce DNA damage (Neri M 2006, Nawrot T 2009) as well as disturb its repair (Jin YH 2003).

Some elements like Fe, Zn, Cu, Co, Cr, Mn, Ni, are needed in small quantities for human metabolism, but may be toxic at higher levels. Others like lead, mercury, cadmium, arsenic etc. have no beneficial role and are positively toxic. Plastics are being increasingly utilized for the storage and delivery of life saving fluids. Although, virgin polymer generally considered being safe due to its inertness, however, the plastic products could be harmful due to migration of certain chemical additives like plasticizers, stabilizers, pigments and unreacted monomers into the stored commodity. Therefore, plastic products intended for storage and delivery of life saving fluids must be evaluated for their safety. In this regards, different countries have laid down various safety assessment tests and guidelines for the suitability and quality assessment of the plastics. To analyze heavy metal contamination in plastic biomedical devices.

## MATERIAL AND METHODS

Four plastic biomedical devices of 21 brands including tubing of intravenous transfusion sets (IV sets, seven brands), dextrose normal saline bottles (DNS bottles, five brands), Ringer lactate bottles (RL bottles, five brands) and Ryle's tubes (four brands) used in the present study were purchased from five major cities of Uttar Pradesh, India, from approved medical shops. Plastic biomedical devices were washed thoroughly with sterilized double distilled water prior to leaching. Aseptic dried plastic biomedical devices were cut into small pieces of 1 $cm^2$ and immersed in 100 ml of either of simulating solvent viz. double distilled water, ethanol (8%), acetic Acid (3%), sodium chloride (0.9%) and sodium carbonate (5%) at 25±2°C for 24h (ambient conditions) and 60±2°C for 2h (elevated conditions). Parallel sets having simulating solvents only were also run under identical conditions and were served as basal control. The leachates will be taken in flask and digested with concentrated Nitric acid and the volume of digested samples will be made upto 10 ml using 1% Nitric acid. The digested samples were analyzed for metal content, (Cd, Cr, Cu, Fe, Mn, Ni, Pb and Zn) using a Perkin- Elmer atomic absorption spectrophotometer. Data were analyzed by one-way analysis of variance (ANOVA) and Dunnett's Multiple Comparison test were employed to assess the significant variations between the control and samples using a computer based software, GraphPad Prism 5. A $p$ value less than 0.05 is considered as significant. Metal content should not be more than 1.00 ppm (Cd should not be more than 0.1 ppm).

## RESULTS AND DISCUSSION

Data were analyzed by one-way analysis of variance (ANOVA) and Dunnett's Multiple Comparison test were employed to assess the significant variations between the control and samples using a computer based software, GraphPad Prism 5. A $p$ value less than 0.05 is considered as significant. The results showed that the leaching of heavy metal in plastic biomedical devices is temperature dependent i.e. high at higher temperature. The heavy metals are not the integral part of the polymers of plastic material. They are used as stabilizer to develop the finished plastic products. So, as with the increase of temperature, plastic molecules would have been received an increased kinetic energy, so the metals attached to the surface or bounded loosely may leave the surface and percolate in the simulating solvents. This might be one among the reasons for higher leaching of metals at higher temperature. At 60±2°C for 2h, the highest mean concentration of metals (cadmium, 0.057±0.002 from IV sets in 0.9% sodium chloride solution; chromium, 0.122±0.002 from IV sets in 5% sodium carbonate solution; copper, 0.138±0.004 from Ryle's tube in 0.9% sodium chloride solution; Iron, 7.476±0.003 from IV sets in 3% acetic acid solution; nickel 0.313±0.02 from RL bottles in 5% sodium carbonate solution; lead 0.220±0.002 from IV sets in 0.9% sodium chloride solution; zinc, 5.954±0.009 from IV sets in 3% acetic acid solution, while in case of double distilled water, 8% ethanol and 3% acetic acid it was not detectable; (Table 1-5). At 25±2°C for 24h, the highest mean concentration of metals (cadmium, 0.051±0.001 from IV sets in 0.9% sodium chloride solution; chromium, 0.121±0.009 from IV sets in 5% sodium carbonate solution; copper, 0.131±0.002 from Ryle's tube in 0.9% sodium chloride solution; iron, 2.41±0.001 from IV sets in 3% acetic acid solution; nickel 0.31±0.004 from RL bottles in double distilled water; lead 0.20±0.001 from IV sets in 0.9% sodium chloride solution; zinc, 1.91±0.002 from IV sets in 3% acetic acid solution (Fig. 8-14). Results showed that the metal concentration were in samples in descending order of iron > zinc > nickel > lead > copper > chromium > cadmium. Leaching was found to be higher at 60±2°C for 2 hours than 25±2°C for 24 hours. All the readings were well within the permissible limits except few. In this study at 60±2°C for 2h, the highest mean concentration of cadmium was observed in the leachates of 0.9% sodium chloride in IV sets (0.057±0.002 ppm). There were no significant difference among the control and samples for cadmium ($P > 0.05$). Highest mean concentration of chromium was observed in the leachates of 5% sodium carbonate in IV sets (0.122±0.002 ppm). There were significant difference among the control and samples for chromium ($P < 0.05$). Highest mean concentration of copper was observed in the leachates of 0.9% sodium chloride in Ryle's tube (0.138±0.004 ppm). There were no significant difference among the control and samples for copper ($P > 0.05$). Highest mean concentration of iron was observed in the leachates of 3% acetic acid in IV sets (7.476±0.003 ppm). There were no significant difference among the control and samples for iron ($P > 0.05$). Highest mean concentration of nickel was observed in the leachates of double distilled water in RL bottles (0.31±0.004 ppm). There were significant difference among the control and samples for nickel ($P < 0.05$). Highest mean concentration of lead was observed in the leachates of 5% sodium carbonate in RL bottles (0.22±0.02 ppm). There were no significant difference among the control and samples for lead ($P > 0.05$). Highest mean concentration of zinc was observed in the leachates of 3% acetic acid in IV sets (5.95±0.009 ppm). There were no significant difference among the control and samples for zinc ($P > 0.05$). At 25±2°C for 24h, the highest mean concentration of metals (cadmium, 0.051±0.001 from IV sets in 0.9% sodium chloride solution; chromium, 0.121±0.009 from IV sets in 5% sodium carbonate solution; copper, 0.131±0.002 from Ryle's tube in 0.9% sodium chloride solution; iron, 2.41±0.001 from IV sets in 3% acetic acid solution; nickel 0.31±0.004 from RL bottles in double distilled water; lead 0.20±0.001 from IV sets in 0.9% sodium chloride solution; zinc, 1.91±0.002 from IV sets in 3% acetic acid solution (Table 1-8). There were no significant difference among the control and samples for all the metals except chromium and nickel. The seven metals (Cd, Cr, Cu, Fe, Ni, Pb and Zn) studied in the present investigation are considered as hazardous material and can severely affect the human health. Results of our study showed that the metal concentration were in samples in descending order of iron > zinc > nickel > lead > copper > chromium > cadmium. There were no significant difference between mean metal concentrations of control and samples for all metal except chromium and nickel. At 60±2°C for 2h concentration of Cd, Cr, Cu, Fe, Mn, Ni, Pb and Zn in the leachates, F =1.569 and $p > 0.05$ for Cd; F = 5.572 and $p < 0.05$ for Cr; F =1.842 and $p > 0.05$ for Cu; F =0.9672 and $p > 0.05$ for iron; F = 0.9882 and $p > 0.05$ for Mn; F = 3.394 and $p < 0.05$ for Ni; F = 2.314 and $p > 0.05$ for lead; F =1.226 and $p > 0.05$ for Zn. At 25±2°C for 24h concentration of Cd, Cr, Cu, Fe, Mn, Ni, Pb and Zn in the leachates, F =1.622 and $p > 0.05$ for Cd; F = 5.625 and $p < 0.05$ for Cr; F =1.167 and $p > 0.05$ for Cu; F =0.9397 and $p > 0.05$ for iron; F = 0.9949 and $p > 0.05$ for Mn; F = 3.705 and $p < 0.05$ for Ni; F = 2.715 and $p > 0.05$ for lead; F =1.642 and $p > 0.05$ for Zn. The results showed that the leaching of heavy metal in plastic biomedical devices is temperature dependent i.e. high at higher temperature. The results clearly revealed that concentration of the inorganic elements used mostly as additives to the plastic biomedical devices. Heavy metals such as Cd, Cr, Cu, Fe, Mn, Ni, Pb and Zn which were found within permissible limit except few according to BIS and International guidelines.

**Significance of Work:** The heavy metals are not the integral part of the polymers. They are used as stabilizer to develop the finished plastic products. Under physicochemical test, Analysis of heavy metals are the gold standard endpoints commonly used to analyze the quality of plastic and polymeric products and based on the quantification of these endpoints, the intended use of that particular plastics is decided.

**Table 1:** Concentration of Cadmium (ppm) in various leachates. The results were reported as a mean ± SD from three set of experiments.

| **Leachates** | **IV sets** | | **DNS bottles** | | **RL bottles** | | **Ryle's tube** | |
|---|---|---|---|---|---|---|---|---|
| | **60±2°C for 2h** | **25±2°C for 24h** | **60±2°C for 2h** | **25±2°C for 24h** | **60±2°C for 2h** | **25±2°C for 24h** | **60±2°C for 2h** | **25±2°C for 24h** |
| DW | ND | ND | ND | ND | ND | ND | ND | ND |
| 8% ethanol | ND | ND | ND | ND | ND | ND | ND | ND |
| 3% acetic acid | ND | ND | ND | ND | ND | ND | ND | ND |
| 0.9% sodium chloride | 0.057± 0.002 | 0.051± 0.001 | ND | ND | ND | ND | ND | ND |
| 5% sodium carbonate | 0.032± 0.009 | 0.031± 0.008 | 0.015± 0.003 | 0.013± 0.003 | 0.014± 0.003 | 0.012± 0.003 | 0.015± 0.003 | 0.013± 0.003 |

**Table2**: Concentration of Chromium (ppm) in various leachates. The results were reported as a mean ± SD from three set of experiments.

| **Leachates** | **IV sets** | | **DNS bottles** | | **RL bottles** | | **Ryle's tube** | |
|---|---|---|---|---|---|---|---|---|
| | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h |
| DW | 0.042± 0.0008 | 0.039± 0.007 | 0.042± 0.001 | 0.035± 0.003 | ND | ND | 0.031± 0.001 | 0.028± 0.001 |
| 8% | ND | ND | 0.019± | 0.010± | ND | ND | ND | ND |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| ethanol | | | 0.001 | 0.003 | | | | |
| 3% acetic acid | 0.103± 0.002 | 0.101± 0.002 | 0.020± 0.001 | 0.017± 0.001 | ND | ND | 0.069± 0.0007 | 0.052± 0.005 |
| 0.9% sodium chloride | 0.085± 0.001 | 0.081± 0.004 | ND | ND | ND | ND | 0.085± 0.002 | 0.082± 0.003 |
| 5% sodium carbonate | 0.122± 0.002 | 0.121± 0.009 | 0.038± 0.001 | 0.032± 0.003 | ND | ND | 0.018± 0.001 | 0.015± 0.008 |

**Table3**: Concentration of Copper (ppm) in various leachates. The results were reported as a mean ± SD from three set of experiments.

| **Leachates** | **IV sets** | | **DNS bottles** | | **RL bottles** | | **Ryle's tube** | |
|---|---|---|---|---|---|---|---|---|
| | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h |
| DW | 0.024± 0.001 | 0.021± 0.001 | 0.005± 0.001 | 0.005± 0.002 | 0.0001± 0.0006 | 0.0001± 0.001 | ND | ND |
| 8% ethanol | 0.050± 0.001 | 0.021± 0.009 | ND | ND | ND | ND | 0.029± 0.0007 | ND |
| 3% acetic acid | 0.080± 0.004 | 0.070± 0.004 | ND | ND | ND | ND | 0.004± 0.003 | 0.003± 0.002 |
| 0.9% sodium chloride | 0.003± 0.001 | 0.002± 0.006 | ND | ND | ND | ND | 0.138± 0.004 | 0.131± 0.002 |
| 5% sodium carbonate | ND | ND | 0.003± 0.001 | 0.002± 0.002 | 0.0002± 0.001 | ND | 0.009± 0.004 | 0.007± 0.003 |

**Table4**: Concentration of Iron (ppm) in various leachates. The results were reported as a mean ± SD from three set of experiments.

| **Leachates** | **IV sets** | | **DNS bottles** | | **RL bottles** | | **Ryle's tube** | |
|---|---|---|---|---|---|---|---|---|
| | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h |
| DW | 0.194± 0.007 | 0.191± 0.001 | 0.323± 0.013 | 0.298± 0.004 | ND | ND | 0.094± 0.008 | 0.085±0.002 |
| 8% ethanol | ND | ND | ND | ND | ND | ND | ND | ND |
| 3% acetic acid | 7.476± 0.003 | 2.41±0.001 | 0.325± 0.013 | 0.321± 0.003 | 0.223± 0.002 | 0.221± 0.001 | 0.321± 0.066 | 0.311±0.006 |
| 0.9% sodium chloride | 0.068± 0.002 | 0.061± 0.006 | 0.224± 0.002 | 0.221± 0.01 | 0.240± 0.012 | 0.231± 0.004 | 0.313± 0.020 | 0.311±0.002 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 5% sodium carbonate | 0.103± 0.002 | 0.101± 0.001 | 0.100± 0.002 | 0.09±0. 001 | 0.241± 0.012 | 0.235± 0.004 | ND | ND |

**Table5**: Concentration of manganese (ppm) in various leachates. The results were reported as a mean ± SD from three set of experiments.

| Leachates | IV sets | | DNS bottles | | RL bottles | | Ryle's tube | |
|---|---|---|---|---|---|---|---|---|
| | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h |
| DW | ND | ND | ND | ND | ND | ND | ND | ND |
| 8% ethanol | ND | ND | ND | ND | ND | ND | ND | ND |
| 3% acetic acid | 0.0005 ±0.001 | ND | 0.142±0.0 04 | 0.131±0.0 01 | ND | ND | 0.002±0.0 01 | 0.001±0.004 |
| 0.9% sodium chloride | ND | ND | ND | ND | ND | ND | ND | ND |
| 5% sodium carbonate | ND | ND | ND | ND | ND | ND | ND | ND |

**Table6**: Concentration of Nickel (ppm) in various leachates. The results were reported as a mean ± SD from three set of experiments.

| Leachates | IV sets | | DNS bottles | | RL bottles | | Ryle's tube | |
|---|---|---|---|---|---|---|---|---|
| | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h |
| DW | 0.001± 0.001 | ND | 0.218± 0.002 | 0.210± 0.003 | 0.051± 0.001 | 0.041± 0.004 | 0.026±0. 001 | 0.021±0.001 |
| 8% ethanol | 0.082± 0.002 | 0.001± 0.002 | 0.221± 0.002 | 0.21±0 .001 | 0.050± 0.002 | 0.048± 0.003 | 0.082±0. 001 | 0.021±0.001 |
| 3% acetic acid | 0.069± 0.001 | 0.057± 0.005 | 0.057± 0.002 | 0.046± 0.001 | 0.056± 0.002 | 0.048± 0.003 | 0.021±0. 003 | 0.018±0.002 |
| 0.9% sodium chloride | 0.033± 0.002 | 0.031± 0.003 | 0.030± 0.001 | 0.029± 0.003 | 0.031± 0.001 | 0.028± 0.001 | 0.021±0. 002 | 0.021±0.004 |
| 5% sodium carbonate | 0.004± 0.001 | 0.003± 0.001 | 0.221± 0.002 | 0.211± 0.002 | 0.313± 0.020 | 0.310± 0.005 | 0.048±0. 001 | 0.039±0.002 |

**Table7**: Concentration of Lead (ppm) in various leachates. The results were reported as a mean ± SD from three set of experiments.

| Leachates | IV sets | | DNS bottles | | RL bottles | | Ryle's tube | |
|---|---|---|---|---|---|---|---|---|
| | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h |
| DW | ND | ND | ND | ND | 0.089±0. | 0.011±0. | ND | ND |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | | | | 005 | 003 | | |
| 8% ethanol | ND | ND | ND | ND | ND | ND | ND | ND |
| 3% acetic acid | 0.017±0.004 | 0.013±0.001 | ND | ND | ND | ND | ND | ND |
| 0.9% sodium chloride | 0.220±0.002 | 0.20±0.001 | ND | ND | ND | ND | 0.046±0.003 | 0.041±0.003 |
| 5% sodium carbonate | 0.199±0.004 | 0.190±0.001 | ND | ND | ND | ND | ND | ND |

**Table8**: Concentration of Zinc (ppm) in various leachates. The results were reported as a mean ± SD from three set of experiments.

| Leachates | IV sets | | DNS bottles | | RL bottles | | Ryle's tube | |
|---|---|---|---|---|---|---|---|---|
| | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h | 60±2°C for 2h | 25±2°C for 24h |
| DW | ND | ND | 0.009±0.003 | 0.008±0.003 | ND | ND | 0.145±0.009 | 0.141±0.003 |
| 8% ethanol | 0.292±0.01 | 0.210±0.001 | ND | ND | ND | ND | ND | ND |
| 3% acetic acid | 5.954±0.009 | 1.91±0.004 | ND | ND | ND | ND | 0.041±0.071 | 0.038±0.03 |
| 0.9% sodium chloride | 0.025±0.004 | 0.020±0.002 | ND | ND | ND | ND | 0.092±0.005 | 0.090±0.003 |
| 5% sodium carbonate | 0.247±0.012 | 0.241±0.003 | ND | ND | ND | ND | 0.022±0.006 | 0.021±0.001 |

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