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COMPARATIVE EVALUATION OF HEAVY METALS IN MARKETED ANTI-DIABETIC CHURNA BY ATOMIC ABSORPTION SPECTROSCOPY

Sanjay K. Bais*[1] and A.V. Chandewar[2]

[*1]Research Scholar, PRIST University, Thanjavur, Tamilnadu, 613 403, India.
[2]P Wadhwani College of Pharmacy, Yavatmal, (MS)-445 001, India.

ABSTRACT

The determination of trace elements in Anti-diabetic churna was carried out by Atomic Absorption Spectrometry. Anti-diabetic churna is herbal medicine used for the treatment of Diabetic mellitus. Marketed Samples of different manufacturer were collected. Heavy metals contents in the investigated samples were found at different levels. The analytical results obtained for all metals indicate that they were present at concentration well below the acceptable daily intake recommended by the World Health Organization. The main purpose of the investigation was to document evidence for the users, and practitioners of marketed Anti-diabetic churna. WHO, (1998) mentions maximum permissible limits in raw materials only for arsenic, cadmium, and lead, which amount to 1.0, 0.3, and 10 p.p.m. respectively. It was found that Arsenic content in which was below the Permissible limit in all formulations. The Cadmium content in AD2 (0.6 p.p.m), AD3 (0.7 p.p.m), AD4 (0.8 p.p.m), AD5 (0.65 p.p.m), AD7 (0.78 p.p.m), AD9 (0.75 p.p.m) and AD10 (0.34 p.p.m) which was above the permissible limits. The lead content in AD2 (13.5 p.p.m), AD5 (12.7 p.pm), AD6 (12.9 p.p.m), AD7 (11.8 p.p.m) and AD9 (14.9 p.p.m) which was above the permissible limits. Such formulations are injurious to health of patient if consumed regularly. Based on the results obtained in the present work, it is concluded that the present techniques are suitable for the routine determination of heavy metals concentration in Anti-Diabetic churna.

KEYWORDS

Anti-diabetic churna, AAS, Metal content, Cadmium and Lead.

Author for correspondence:
Sanjay K. Bais,
Research Scholar, PRIST University,
Thanjavur-Tamilnadu-613403,
India.

Email: sanjaybais@rediffmail.com

INTRODUCTION

Herbs and products containing herb(s) have been in trade and commerce and are currently used for a variety of purposes[1]. The WHO defines an herb as being fresh or dried, fragmented or powdered plant material, which can be used in this crude state or further processed and formulated to become the final herbal product[2]. In India, drugs of herbal origin have been used in traditional systems of medicines such as

Unani and *Ayurveda* since ancient times. The *Ayurveda* system of medicine uses about 700 species, *Unani* 700, *Siddha* 600, Amchi 600 and modern medicine around 30 species[3].

Over the past decade several news-catching episodes in developed communities indicated adverse effects, sometimes life threatening, allegedly arisen consequential to taking of OTC herbal products or traditional medicines from various ethnic groups. These OTC products may be contaminated with excessive or banned pesticides, microbial contaminants, heavy metals, chemical toxins, and for adulterated with orthodox drugs. Excessive or banned pesticides, heavy metals and microbial contaminants may be related to the source of these herbal materials, if they are grown under contaminated environment or during collection of these plant materials. Chemical toxins may come from unfavorable or wrong storage conditions or chemical treatment due to storage. The presence of orthodox drugs can be related to unprofessional practice of manufacturers. Some of these environment related factors can be controlled by implementing standard operating procedures (SOP) leading to Good Agricultural Practice (GAP), Good Laboratory Practice (GLP), Good Supply Practice (GSP) and Good Manufacturing Practice (GMP) for producing these medicinal products from herbal or natural sources. The public's belief that herbal and natural products are safer than synthetic medicines can only be ascertained by imposing regulatory standards on these products that should be manufactured using this Good Practices[4].

The manufacture of the finished products should be in accordance with the good manufacturing practices (GMPs), with post-marketing quality assurance surveillance Evaluation of the toxicity and adverse drug reaction of the herbal preparation has been a neglected area, as herbs are considered natural products and, therefore safe. This lack of information makes it difficult to compare the benefit-risk profile of herbal medicines. Besides, the comparison of traditional medicines with modern drugs with comparative efficacy has not been conducted for most of the drugs[5].

WHO, (1998) mentions maximum permissible limits in raw materials only for arsenic, cadmium, and lead, which amount to 1.0, 0.3, and 10 p.p.m, respectively. The concentration of heavy metals is one of the criteria that make raw plants admissible to the production of medicines due to the fact that amount taken increases with the concentration, increased by constant mass of a taken dose[6].

Herbal medications are claimed and widely believed to be beneficial; however, there have been reports of acute and chronic intoxications resulting from their use. The popularity and availability of the traditional remedies have generated concerns regarding the safety, efficacy and responsibility of practitioners using traditional remedies. A common misperception is that medicaments of natural substances cannot be present in toxic concentrations in a variety of herbal preparations and dietary supplements[7].

Arsenic

Arsenic is a highly toxic, naturally occurring grayish- white element used as a poison in pesticides and herbicides. Arsenic is also found as an ingredient in pigments and wood preservatives. Arsenic contained in wolmanized lumber will not release toxic compounds unless burned. Arsenic can be harmful through inhalation, absorption through skin and mucous membranes, skin contact, and ingestion[8].

Cadmium

Cadmium is a toxic heavy metal, well known for its occupational health risk, and cadmium (as a pollutant of air and water) is an increasing public health concern. Inhalation of cadmium fumes or dust is the primary cause of cadmium exposure[9].

Most studies have centered on the detection of early signs of kidney dysfunction and lung impairment in the occupational setting, and, in Japan, on the detection and screening for bone disease in general populations exposed to cadmium-contaminated rice. More recently, the possible role of cadmium in human carcinogenesis has also been studied in some detail[10].

Lead

Lead is a ubiquitous toxicant. Lead poisoning is an insidious disease that can result in developmental

delays, behavioral disorders and irreversible brain damage. The major signs and symptoms of lead poisoning are pallor, gingival lead line, gastrointestinal disorder, and anemia, renal and neurological symptoms (peripheral neuropathy, ataxia and memory loss) in adults. Chronic exposure to lead is associated with renal dysfunction whilst, chronic lead toxicity will also lead to sterility in adults[11].

Diabetes mellitus

The term diabetes mellitus describes a metabolic disorder of multiple etiologies characterized by chronic hyperglycemia with disturbances of carbohydrate, fat and protein metabolism resulting from defects in insulin secretion, insulin action, or both. The effects of diabetes mellitus include long-term damage, dysfunction and failure of various organs. Diabetes mellitus may present with characteristic symptoms such as thirst, polyuria, blurring of vision, and weight loss. In its most severe forms, ketoacidosis or a non–ketosis hyperosmolar state may develop and lead to stupor, coma and, in absence of effective treatment, death. Often symptoms are not severe, or may be absent, and consequently hyperglycemia sufficient to cause pathological and functional changes may be present for a long time before the diagnosis is made. The long–term effects of diabetes mellitus include progressive development of the specific complications of retinopathy with potential blindness, nephropathy that may lead to renal failure, and/or neuropathy with risk of foot ulcers, amputation, Charcot joints, and features of autonomic dysfunction, including sexual dysfunction. People with diabetes are at increased risk of cardiovascular, peripheral vascular and cerebrovascular disease. Several pathogenetic processes are involved in the development of diabetes. These include processes which destroy the beta cells of the pancreas with consequent insulin deficiency, and others that result in resistance to insulin action. The abnormalities of carbohydrate, fat and protein metabolism are due to deficient action of insulin on target tissues resulting from insensitivity or lack of insulin[12].

ABOUT ANTIDIABETIC CHURNA

Antidiabetic churna is one of the well-known powdered preparations of Indian system of medicine being used in Ayurveda. This is well known phytomedicines is made in combination with the following composition

Neem (Azadirachta Indica) :	20%,
Karela (Momordica Charantia) :	15%
Jamun (Eugenis Jambolana) :	20%,
Amla (Emblica Officinalis) :	10%
Gurmer (Gymnema Sylvestra) :	10%
Glo :	7%,
Paner Dodi :	7%,
Methi (Trigohella Foenum)	qs

This proportion as reported in Ayurvedic Formulary of India (AFI). Anti Diabetic Powder is widely consumed by people affected from Diabetes. Anti Diabetic Powder includes pure extracts of jamun, neem, and karela. It is indicated to reduce sugar level in patients suffering from Diabetes Mellitus avoiding the future complications of Diabetes[13].

MATERIALS AND METHODS

All 10 samples were analyzed for toxic metal contamination

Determination of Heavy Metals in Herbal Formulations

Study Design

An experimental method of research was performed to assess the presence or absence of heavy metals in selected Herbal formulations; and the concentration of each heavy metal was determined by AAS method.

Preparation of Standards

Arsenic standard solution

Arsenic standard solution was prepared from Stock solution (1000 ppm Reagecon stock solution) standard solution of concentrations 0.02, 0.04, 0.06, 0.08 and 0.10 ppm were prepared The absorption of standard solution was measured at 193 nm using hallow cathode lamp as a light source and air acetylene flame with N_2 gas source on Hydride generator mode on Atomic absorption Spectrophometer was shown in Table No.1 and Figure No.1.

Cadmium Standard solution

Cadmium standard solutions were prepared from Stock solution (1000 ppm Reagecon stock solution) standard solution of concentrations 0.2, 0.4, 0.6, 0.8 and 1.0 ppm were prepared. The absorption of standard solution measured at 228.8 nm using hallow cathode lamp as a light source and air acetylene blue flame on Atomic absorption Spectrophometer. The results are expressed in Table No.2 and Figure No.2.

Lead standard solution

Lead standard solutions were prepared from Stock solution (1000 ppm Reagecon stock solution) standard solution of concentrations, 2, 4, 6.8, 10 ppm., were prepared. The absorption of standard solution measured at 217 nm using hallow cathode lamp as a light source and acetylene blue flame on Atomic absorption Spectrophometer expressed in Table No.3 and Figure No.3.

Calibration of Equipment

For the studied elements we established the following sensitivity and detection limits, respectively of the used flame atomic absorption Spectrophotometer (AAS) apparatus. As 0.02 and 0.10 ppm, Cd 0.2 and 1.0 ppm, Pb 2 and 10.0 ppm[14].

Extraction of heavy metals from Herbal formulations

A sample of 10gm of each herbal formulation was taken in a silica crucible and heated to remove the moisture. It was then put in a muffle furnace at 450°C, for 2 hours, to remove the organic material. The ash was digested in 5 ml dilute HCL + 1ml HNO_3, cool, 20 ml distilled water added. Filtered and the filter paper were washed distilled water, in 100ml volumetric flask. It was made to 100ml with distilled water and suitable dilutions were prepared. This filtrate contained the metal-like arsenic, lead cadmium. The Arsenic, cadmium and Lead were determined by Atomic Absorption Sphectrophometer[15].

RESULTS AND DISCUSSION

People generally use herbal medicine for prolonged period of time to achieve desirable effects. Prolong consumption of such herbal medicine might reduce chronic or subtle health hazards. Thus our findings indicate that the medicinal plant or plant parts used for different diseases must be checked for heavy metals contamination in order to make it safe for human consumption. The general belief that herbal preparations are natural and, therefore, inherently safe harmless and without any adverse effects is sometimes unfounded. Toxic effects of herbal preparations have been attributed to several factors including contamination by poisoning through traditional Chinese, Indian and Malaysian medicines have been reported expressed in Table No.4-7 and Figure No.4-7.

Table No.1: Concentration and absorbance of Arsenic

S.No	Solution. No	Concentration	Absorbance
1	1	0.02	0.066
2	2	0.04	0.152
3	3	0.06	0.238
4	4	0.08	0.298
5	5	0.10	0.384

Table No.2: Concentration and absorbance of Cadmium

S.No	Solution. No	Concentration	Absorbance
1	1	0.200	0.034
2	2	0.400	0.051
3	3	0.600	0.080
4	4	0.800	0.106
5	5	1.000	0.129

Table No.3: Concentration and absorbance of Lead

S.No	Solution. No	Concentration	Absorbance
1	1	2.000	0.027
2	2	4.000	0.062
3	3	6.000	0.094
4	4	8.000	0.131
5	5	10.000	0.164

Table No.4: Arsenic content in Anti-Diabetic churna AD1 to AD10

S.No	Formulation code	Arsenic content (ppm)	Remark
1	AD1	0.23	Within permissible limit
2	AD2	0.56	Within permissible limit
3	AD3	0.32	Within permissible limit
4	AD4	0.26	Within permissible limit
5	AD5	0.51	Within permissible limit
6	AD6	0.37	Within permissible limit
7	AD7	0.66	Within permissible limit
8	AD8	0.28	Within permissible limit
9	AD9	0.45	Within permissible limit
10	AD10	0.68	Within permissible limit

Determination of Cadmium Content

Table No.5: Cadmium content in in Anti-Diabetic churna AD1 to AD10

S.No	Formulation code	Cadmium content (ppm)	Remark
1	AD1	BDL	within permissible limit
2	AD2	0.6	Above permissible limit
3	AD3	0.7	Above permissible limit
4	AD4	0.8	Above permissible limit
5	AD5	0.65	Above permissible limit
6	AD6	BDL	within permissible limit
7	AD7	0.78	Above permissible limit
8	AD8	BDL	Above permissible limit
9	AD9	0.75*	Above permissible limit
10	AD10	0.34*	Above permissible limit

BDL= Below detectable limit

Determination of Lead Content

Table No.6: Lead content in in Anti-Diabetic churna AD1 to AD10

S.No	Formulation code	Lead content (ppm)	Remark
1	AD1	7.3	within permissible limit
2	AD2	13.5	Above permissible limit
3	AD3	8.3	within permissible limit
4	AD4	5.8	within permissible limit
5	AD5	12.7	Above permissible limit
6	AD6	12.9	Above permissible limit
7	AD7	11.8	Above permissible limit
8	AD8	1.9	within permissible limit
9	AD9	14.9	Above permissible limit
10	AD10	4.2	within permissible limit

BDL= Below detectable limit

Table No.7: Comparative data of Heavy metals contents in Anti-Diabetic churna AD1 to AD10

S.No	Formulation code	Heavy metal content mg/kg		
		Arsenic	Cadmium	Lead
1	AD1	0.23	BDL	7.3
2	AD2	0.56	0.6	13.5
3	AD3	0.32	0.7	8.3
4	AD4	0.26	0.8	5.8
5	AD5	0.51	0.65	12.7
6	AD6	0.37	BDL	12.9
7	AD7	0.66	0.78	11.8
8	AD8	0.28	BDL	1.9
9	AD9	0.45	0.75*	14.9
10	AD10	0.68	0.34*	4.2

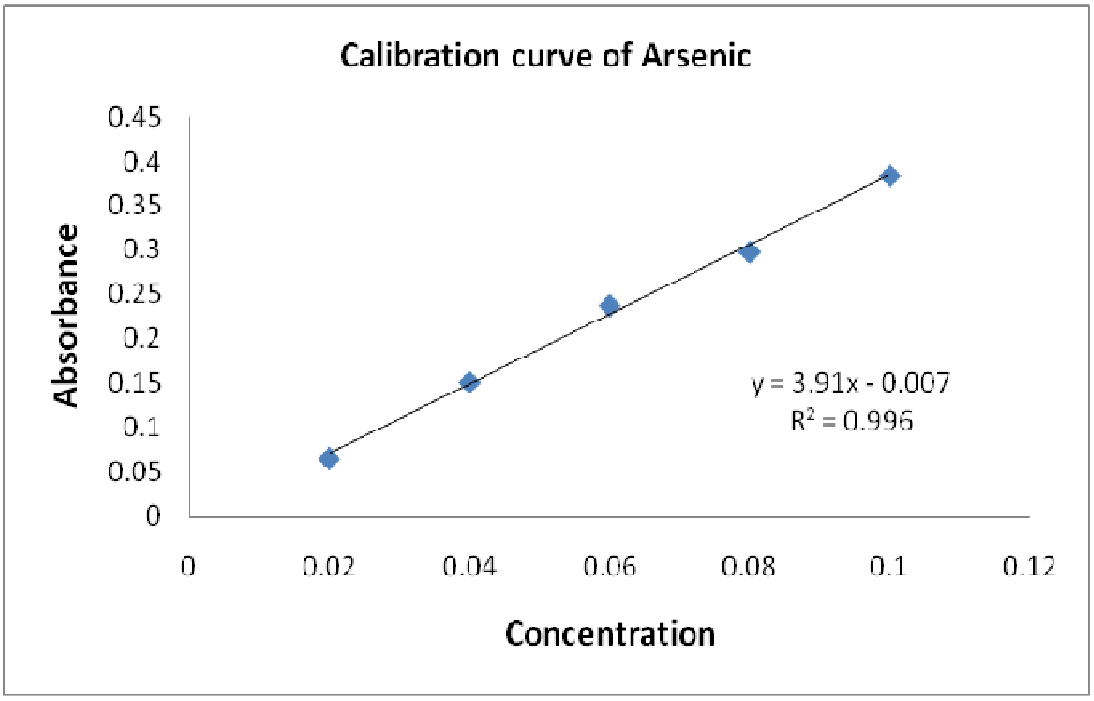

Figure No.1: Calibration Curve of Arsenic

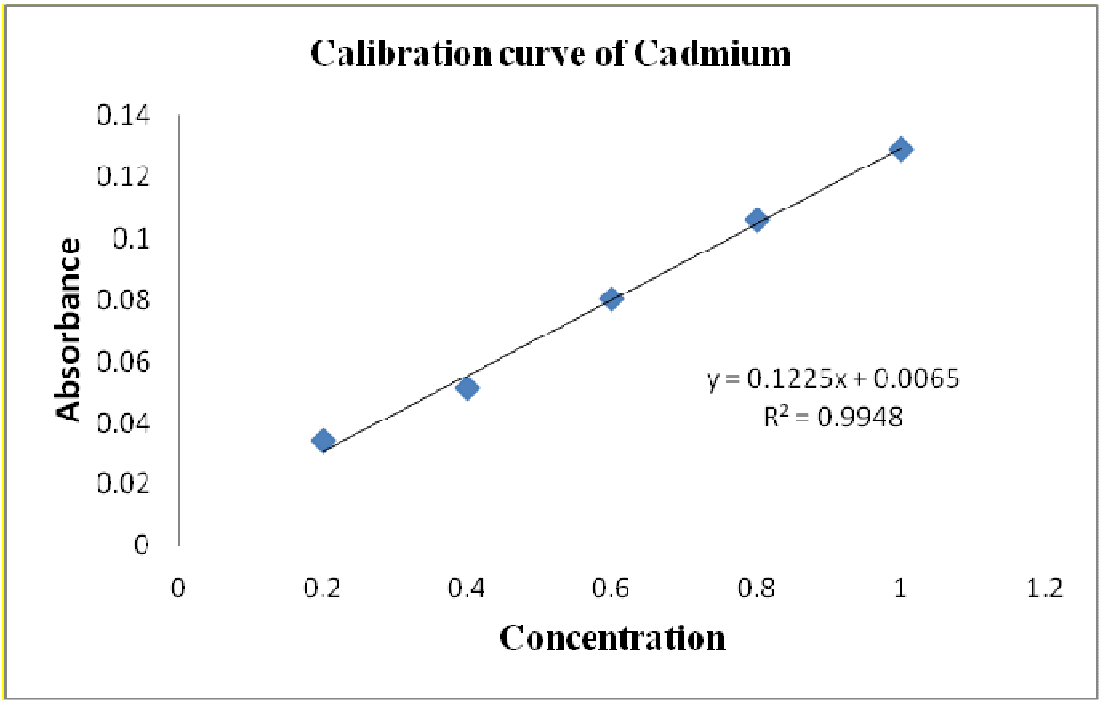

Figure No.2: Calibration curve of Cadmium

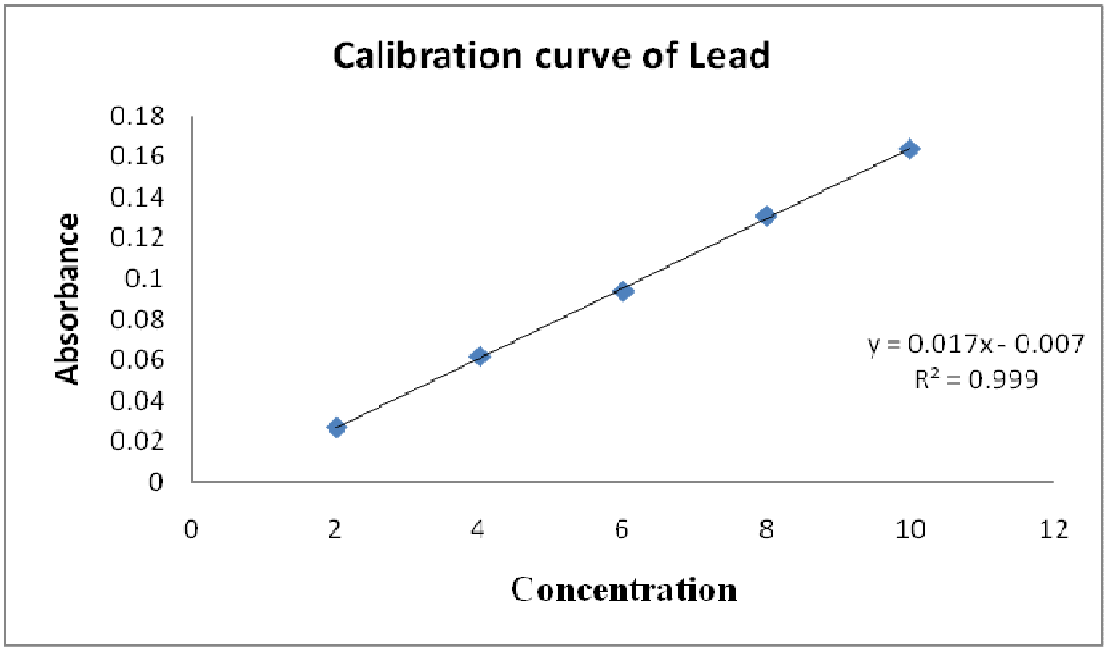

Figure No.3: Calibration curve of lead

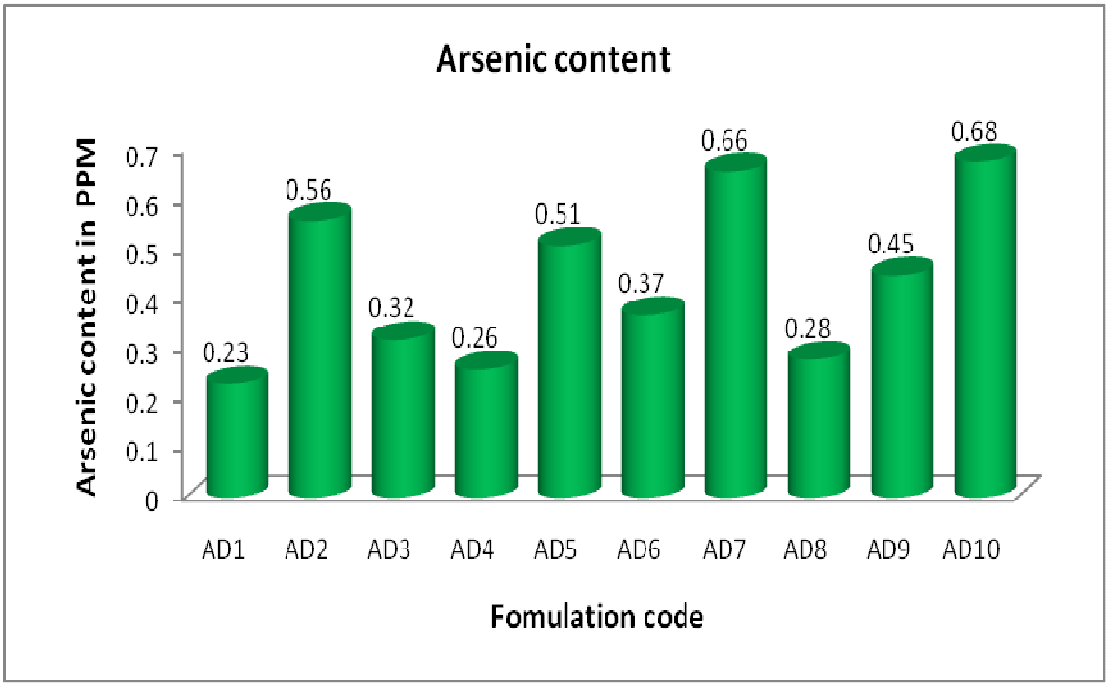

Figure No.4: Arsenic content in Anti-Diabetic churna AD1 to AD10

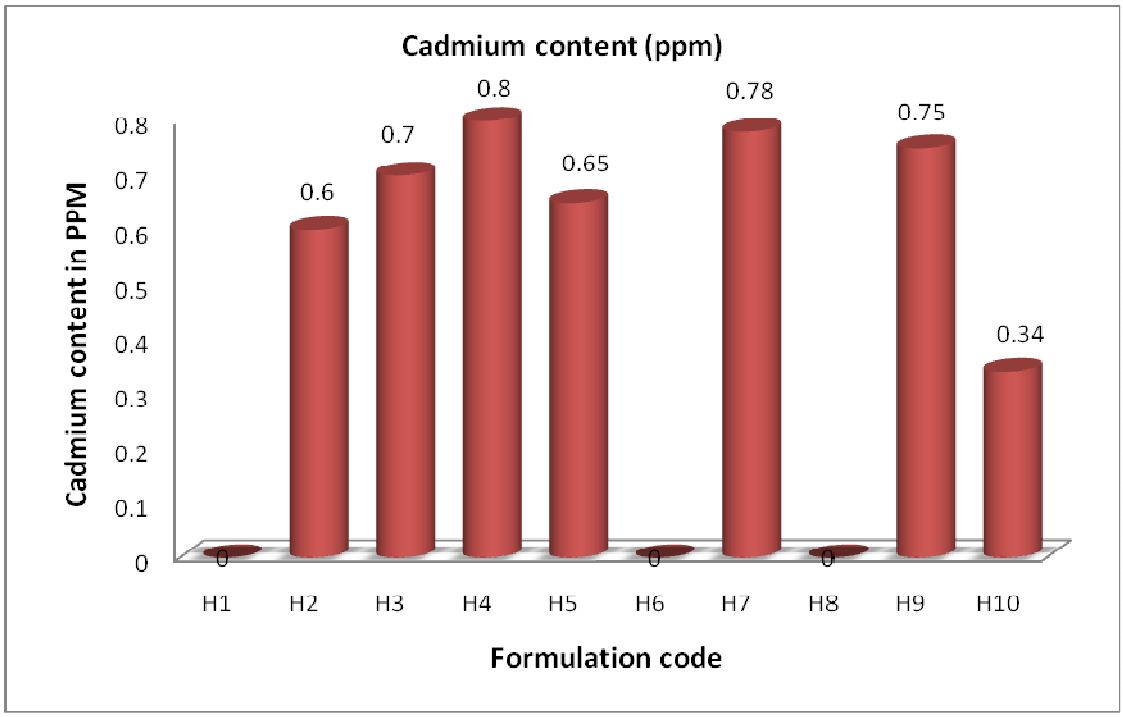

Figure No.5: Cadmium content in Anti-Diabetic churna AD1 to AD10

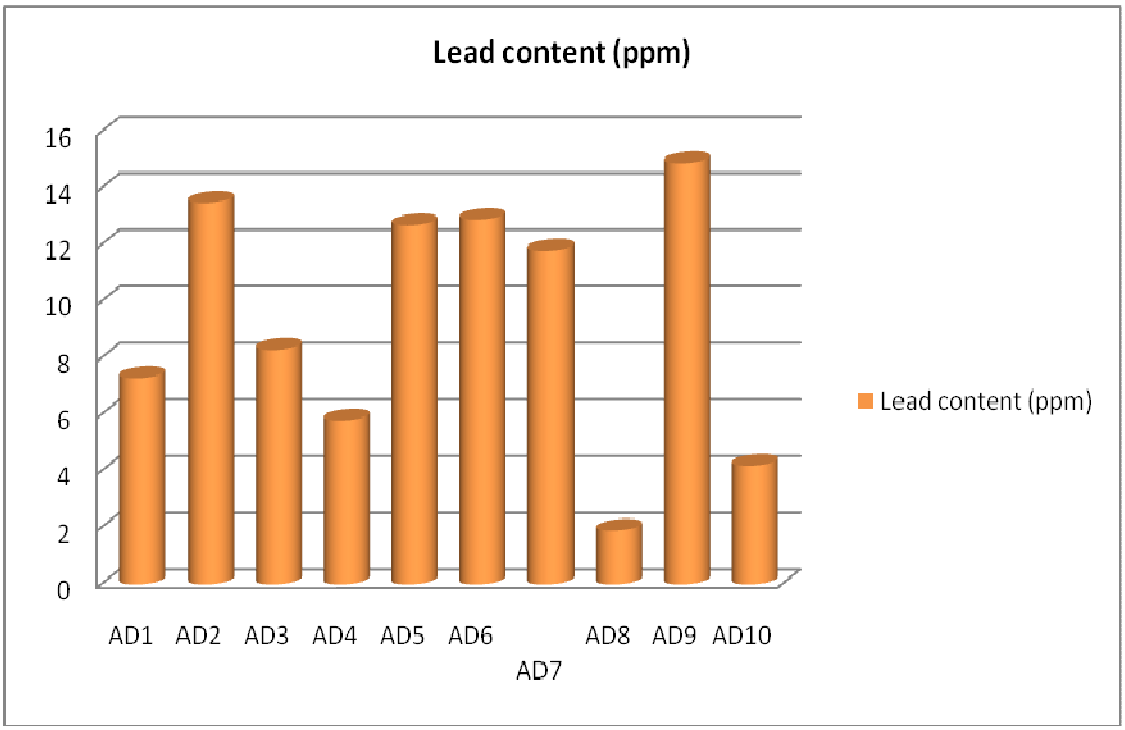

Figure No.6: Lead content in Anti-Diabetic churna AD1 to AD10

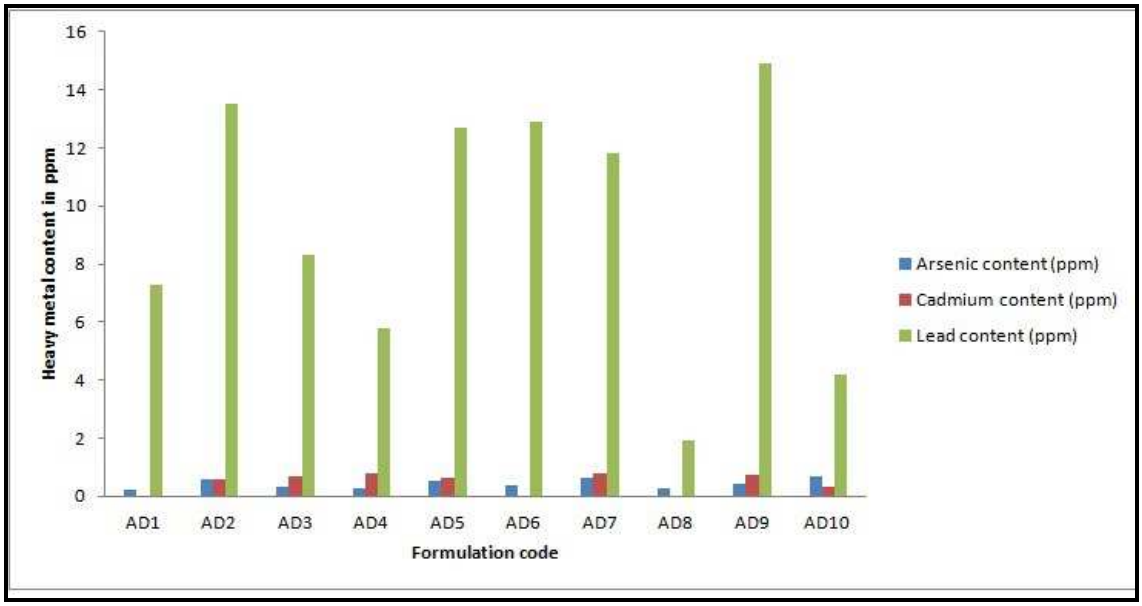

Figure No.7: Comparative data of Heavy metals contents in Anti-Diabetic churna AD1 to AD10

CONCLUSION

WHO, (1998) mentions maximum permissible limits in raw materials only for arsenic, cadmium, and lead, which amount to 1.0, 0.3, and 10 ppm, respectively. The present work indicates that there is presence of Heavy Metal contents in Herbal formulations selected for study. It was found that Arsenic content in marketed Anti-Diabetic churnas was below the Permissible limit in all formulations. The Cadmium content in AD2 (0.6 p.p.m), AD3 (0.7 p.p.m), AD4 (0.8 p.p.m), AD5 (0.65 p.p.m), AD7 (0.78 p.p.m), AD9 (0.75 p.p.m) and AD10 (0.34 p.p.m) which was above the permissible limits. The lead content in AD2, (13.5 ppm), AD5 (12.7 ppm), AD6 (12.9 ppm), AD7 (11.8ppm) and AD9 (14.9 ppm) which was above the permissible limits.

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CONFLICT OF INTEREST

We declare that we have no conflict of interest.

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