CHAPTER 1

INTRODUCTION

In 1950, Minamata bay tragedy caught the world unawares. Minamata Bay was heavily polluted by wastewater, mixed with mercury dumped into Hyakken Harbour from the Chisso Corporation's factory in Minamata, particularly by methylmercury. The highly toxic compound bioaccumulated in fish and shellfish in the bay which, when eaten by the people living around the bay, gave rise to Minamata disease. More than 10,000 people were affected.

It has been since recognized that the multiple pathways of mercury contamination through air, water, food, pharmaceuticals, cosmetic products etc., pose serious concern because it persists in the environment and accumulates in the food web. Amongst three forms of mercury, the organic form is most toxic as it passes through the blood brain barrier owing to its lipid solubility. The damage has vast implications with human beings at the top of food chain getting worst of the deal owing to bio magnification.

1.1 SOURCE OF MERCURY

1.1.1 Mercury in air

As a natural element mercury is ubiquitous in the environment, approximately 10,000 tons originates from degassing of earth's crust, to this amount approximately 20,000 tons/year is added by anthropogenic activity [1]. Mercury emission from the coal smoke is the main source of anthropogenic discharge and mercury pollution in atmosphere. It is estimated that mercury emissions will increase at a rate of 5% a year [2].

When medical devices like thermometer, sphygmomanometer or household items like fluorescent night lamps or thermostats are discarded residual mercury is emitted. The US Environmental Protection Agency (EPA) and National Emissions Inventory (NEI) had
the most complete coverage for all states. It found coal-fired electric utilities accounted for 52.7% of the region's Hg emissions.

Other important contributors to regional emissions included municipal waste combustion (5.6%), mercury cell chloralkali plants and hazardous waste incinerators (4% each), stationary internal combustion engines (ICEs) (3.5%), industrial, commercial and institutional (ICI) boilers (3.3%) and lime manufacturing (3.0%) and medical waste incineration (1%) [3].

Informal gold mining has used mercury since antiquity. High contamination of Brazilian Amazon (Brazil is world's second largest producer of gold) is indicated by the strong presence of mercury in its biota [4]. It is an occupational hazard for dental staff [5], chloralkali factory workers [6] and gold miners etc.

1.1.2 Mercury in water

Mercury in air eventually passes into rivers, lakes and oceans after travelling long distances together with wind. With mercury contaminating rain [7, 8], ground and seawater [9], no one is safe. Mercury cloud water concentrations ranged from 7.5 to 71.8 ng l (−1), with a mean of 24.8 ng l (−1). Liquid water content explained about 60% of the variability in Hg cloud concentrations [10]. There are also linkages between acidic deposition and fish mercury contamination and eutrophication of estuaries [11]. Numerous factories that directly pump untreated effluents pollute groundwater. The polluted water produces acidic rain which ultimately contaminates all water bodies.

Report published in a reputed Indian daily, the hindustan times showed result of water samples analysed at IIT, Kanpur. Ground water samples in India from eight places each from Punjab, Haryana, Andhra Pradesh, Gujarat and Kanpur showed surprisingly high levels of Hg in all samples.

Water sample from Panipat (Haryana) had highest level of Hg at concentration 268 times that of safe limit, even the sample with least Hg value had 58 times more mercury than the upper safe limit. Algal bloom and leaf fall events can result in elevated methyl mercury (MeHg) concentrations in surface waters, potentially leading to increased MeHg accumulation in fish [12].
1.2 Mercury contamination of food

1.2.1 Food of animal origin

The emitted mercury both natural and anthropogenic is in an inorganic form predominantly as metallic vapour, which is carried to great distances by winds and eventually falls in water bodies. In aquatic environments, inorganic mercury is microbiologically transformed into lipophilic organic compound, methyl mercury. This transformation makes mercury more prone to bio magnification in food chains. Consequently, populations with traditionally high dietary intake of food originating from fresh or marine environment have highest dietary exposure to Hg.

Extensive research done on locals across the globe have already established this for instance polar Eskimos. People who routinely consume fish or a particular species (Mackerel King) of fish are at an increased risk of methyl mercury poisoning [1]. Since mercury intake is expressed on a per kilogram body weight basis exposure of children under age 14 is two-three times high because of higher food intake per kilogram body weight.
After measuring total mercury in the edible portions of 244 selected fishes and shellfish purchased in Canada at retail level, the Canadian advisory to children and women of child bearing age is to limit their consumption of fresh and frozen tuna, swordfish and shark to no more than one meal per month [13].

When 21 fish species, cephalopods and crustaceans were analysed for mercury accumulation the former two ranked highest [14]. In Yokon delta system, bio magnification factor of 12 was calculated for methyl mercury, out of 29% fish species, 62% contained Hg exceeding wildlife critical value for piscivorous animals. Overall 24% fish exceeded critical value for human consumption and 58% wildlife critical value [15]. Cattle and pigs kept in area with contaminated river water had twice the concentration of blood and hair Hg than control ones [16].

1.2.2 Food of plant origin

Emissions of mercury from the province of Guizhou in Southwestern China to the global atmosphere have been estimated to be approximately 12% of the world total anthropogenic emissions primarily due to mining, chemical discharge and electricity production. Even though the major source of mercury is inorganic, it was observed that active transformation of inorganic mercury to organic mercury species (MeHg) takes place in water, sediments and soils. It has been reported that the concentration of mercury in rice grains can reach up to 569 g/kg of total Hg of which 145 g/kg is in MeHg form [17]. While analysing in situ aquatic and terrestrial plants in vicinity of chloralkali plants growing at Hg conc. 8.9 mg/kg it was found that Cabbage, Bracica oleracea and Amaranthus oleraceous accumulated mercury at significant levels [18]. Amongst edible mushrooms representing eight species, the highest average content of mercury was found in Boletus pinicola at 7.37 ppm DW [19]. In south east Asia, the aquatic macrophyte water spinach (Ipomoea aquatica Forsk) is a popular vegetable that is cultivated in freshwater sources, it was found that the vegetable accumulated various heavy metals like mercury, cadmium and lead in a nutrient deficient medium [20].
1.3 Mercury in pharmaceuticals and utility products

Mercury has always been a popular choice for dental amalgams. Thimerosol is a mercury containing compound used as a preservative in Hepatitis B, Diphtheria, Pertussis, Acellular pertussis and Tetanus vaccines. Use of mercury in vaccines have caused furore in concerned circles owing to death of infants and speculations over long-term effects [21]. Infants are exposed to phenyl mercury from treated diapers and young children ingesting mercuric chloride in teething powders have been found to develop acrodynia and kawasaki disease [22]. Skin whitening creams and soaps from developing countries is a recognized source of chronic mercury poisoning [23,24].

1.4 Mercury and wildlife

It is well known that heavy metals in larger amounts are toxic to animals as well as plants; and mercury is no exception to this, though some of these metals may actually be required in trace amounts to support life. The general signs and symptoms of mercury toxicity for sheep, cattle, pig, chicken and turkey include lack of appetite, loss of weight, muscular incoordination, unstable gait and lameness. Sea birds from mercury contaminated colony, metal dosed birds and metal dosed mice have demonstrated nephrotoxic lesions of severe type [25]. The possible effects of heavy metal exposure on the condition and health of great tit nestlings (Parus major) at four study sites along a pollution gradient near a large nonferrous smelter in Belgium during three consecutive breeding seasons was taken. When taking into account the number of young in the nest at the time of sampling, nestling body mass and condition were significantly reduced at the most polluted site [26]. Methyl mercury was attributed for decrease in reproduction of adult fat head minnows at dietary concentrations encountered by predatory fishes in aquatic systems with contaminated food webs, implying that exposed fish populations could be adversely affected by this widespread contaminant [27]. Inorganic mercury disturbs a part of respiration process in shrimp larvae Pandalus borealis [28]. Embryo toxicity and teratogenicity of organic mercury compounds have been observed in fish, birds and even mammals [29].
**1.5 HERBAL MEDICINE**

Over the past decade, herbal medicine has become a topic of global importance, making an impact on both world health and international trade. Medicinal plants continue to play a central role in the healthcare system of large proportions of the world’s population [30].

The history reveals that medicinal plants have traditionally served as man’s most important weapon against pathogens. People use traditional, natural remedies for curing different types of diseases and even in the next millennium the herbal medical practice will forge as important remedy to treat various disorders. This is particularly true in developing countries, where herbal medicine has a long and uninterrupted history of use. Recognition and development of the medicinal and economic benefits of these plants are on the increase in both developing and industrialized nations [31]. Continuous usage of herbal medicine by a large proportion of the population in the developing countries is largely due to the fact that herbal medicines are more acceptable in these countries from their cultural and spiritual points of view and the high cost of Western pharmaceuticals and healthcare [32].

Medicinal plants are treated as a subject of serious study and are undergoing intense research all over the world. India is known as the emporium of medicinal plants due to the occurrence of several thousands of medicinal plants in the different bioclimatic zones. In India, the use of different parts of several medicinal plants to cure specific ailments has been in vogue from ancient times.

The Ayurveda and Siddha system of medicines are the traditional heritage of India which has been in existence for several centuries which cater to the needs of nearly seventy per cent of our population residing in villages. These systems include many time tested medicinal plants or drugs for various diseases and to which there is no answer in modern medicine till today.
Age-old traditions utilized plants and animals either in their native form or their crude extracts for various therapeutic effects. Even though many modern medicines are either derived or modelled based on compounds found in nature, the use of natural drugs has declined in last three decades. The current trend is witnessing a rapid change in this scenario where for a variety of reasons more individuals nowadays prefer to take personal control over their health with the use of herbal medicines, not only in the prevention of diseases but also to treat them. This is particularly true for a wide variety of illness readily treated at home (common cold etc..) [33]. Herbal products are also commonly used by patients with certain chronic medical conditions including breast cancer (12%), liver disease (21%), human immunodeficiency virus (22%), asthma (24%), and rheumatological disorders (26%). Even as we commence the new century with its exciting prospect of gene therapy, herbal medicines remain as one of the common forms of therapy available to the world population [34].

Herbal based medicines do have advantages over chemical compounds. Active constituents in plants are always biologically balanced, affect the human organism in a complex manner, do not usually accumulate in the body and are capable of neutralizing the harmful effects of chemical compounds. It is also noteworthy that some of the most important drugs of past 50 years or so, which have revolutionized modern medical practices, have almost all first been isolated from plants. These wonder drugs include curare alkaloids, penicillin, vincoleucoblastin, podophyllotoxin, atropine, digitoxin, stropanthidin and other new therapeutic agents. Natural products may also be used as building blocks for the synthesis of semi synthetic drugs like plant saponins which can be extracted and easily altered chemicals to produce sapogenins for the manufacture of steroidal drugs [35].

1.5.1 IMPORTANCE OF NATURAL PRODUCTS

1. They provide a number of extremely useful drugs that are difficult to produce commercially by synthetic means (e.g. Ergot, Digitalis).

2. Natural resources also supply basic compounds that may be modified slightly to render them more effective or less toxic.
3. Natural products are used as prototypes or models for synthetic drugs possessing physiological activities similar to the original drug [36].

4. As a source of new lead compounds of novel chemical structure.

1.5.2 CURRENT PROBLEMS FACED BY HERBAL MEDICINE

India is sitting on a gold mine of well-recorded and well practiced knowledge of traditional herbal medicine. Unlike China, India has not been able to capitalize on this herbal wealth by promoting its use in the developed countries despite their renewed interest in herbal medicines. This can be achieved by judicious product identification based on diseases found in the developed countries for which no medicine or only palliative therapy is available; such herbal medicines will find speedy access into those countries. Strategically, India should enter through those plant-based medicines which are already well accepted in Europe, USA and Japan. Simultaneously, it should identify those herbs (medicinal plants) which are time-tested and dispensed all over India. The basic requirements for gaining entry into developed countries include: (i) well-documented traditional use (ii) single plant medicines (iii) medicinal plants free from pesticides, heavy metals etc., (iv) standardization based on chemical and activity profile (v) safety and stability. However, mode of action studies in animals and efficacy in human will also be supportive. Such scientifically generated data will project herbal medicine in a perspective and help in sustained global market [37].

1.5.3 HERBAL MEDICINE SCENARIO IN INDIA

Ours is a vast country where wide variations in climate, soil, altitude and latitude are available. Nature has bestowed on us a very rich botanical wealth and a large number of diverse types of plants growing wild in different parts of the country. The people of India had an incredible knowledge of phytomedicine driven apparently by the tremendous passion for the study and use of medicinal plants. Indians have thus produced one of the world’s richest medicinal plant heritage. The Western Ghats have been designated as one of the hot spots of global biodiversity. A rich depository of flora with high endemism is found in the Himalayas. The immense taxonomic diversity of the country throws a challenge to the Indian chemists and biologists to transform the enormous bio resource into
economic wealth and intellectual property. The state Tamil Nadu is endowed with a very rich flora due to the various physiographic features and physiognomic factors, different types of vegetation exist in the state. A total of 5640 species of the flowering plants (including 6 gnomosperms) are reported either naturally occurring or cultivated in the state [38]. Out of this 1474 have been reported to be medicinal plants. However, our knowledge of medicinal plants has mostly been inherited traditionally. Use of plants for curing various ailments is not confined to the doctors only but is known to several households as well. The use of bioactive plant derived compounds is on the rise in the world because natural remedies are somehow safer and more efficacious than the remedies that are pharmaceutically derived which are more dangerous than the diseases [39,40]. On the contrary the plant derived medicines are based upon the premise that they contain natural substances that can promote health and alleviate illness. So a retrospection of the healing power of the plants and the return to natural remedies is an absolute need of our time. However a detailed investigation and documentation of plants used in local health traditions and ethno pharmacological evaluation to verify their efficacy and safety is very essential which can lead to the development of new valuable herbal drug for the treatment of various diseases. In view of the wide spread interest on the Smilax china, the present study have been undertaken for the protective activity against Mercuric chloride intoxication.

The genus Smilax, belongs to the Liliaceae family, contains 350 species, and is widely distributed in the tropical and temperate zones throughout the world, and especially in tropical regions of East Asia, and South and North America [41]. Many of them have been used as medicinal herbs in East Asian countries. For example, Smilax china L. is commonly used in traditional Chinese medicine as diuretic, for treatment of rheumatic arthritics, detoxication, lumbago, gout, tumor, inflammatory diseases and for detoxification [42].

Recent pharmacological investigations showed that Smilax china has anti-inflammatory activity [43-46] and some steroidal saponins isolated from this plant exhibited significant cytotoxicity against several tumor cell lines [47,48]. Furthermore, kaempferol-7-O-[β]-D-glucoside isolated from Smilax china rhizome induces apoptosis on cancer cells [49].
Oxidative metabolism is essential for normal biological activities of cell. However, it also accompanies the production of reactive oxygen species (ROS) [50]. Oxidative modification of DNA, proteins, lipids, and small cellular molecules by excess of ROS plays a role in a wide range of common diseases such as cardiovascular diseases, neurodegenerative disorders, cancer, and aging. Oxidation also affects food quality. It is a major cause for food deterioration through affecting color, flavor, texture, and nutritional value.

Antioxidants are vital substances which possess the ability to reduce oxidative damage caused by ROS [51,52]. Due to the possible carcinogenic effects of synthetic antioxidants, it has been suggested that natural antioxidants are more safe and healthy than synthetic antioxidants when used in food [53,54]. In order to scientifically evaluate the folklore claims this plant has been selected for the present investigation in view of its therapeutic activities against Mercuric Chloride intoxication.