CHAPTER - 1

1. REVIEW OF LITERATURE

1.1. CADMIUM IN THE ENVIRONMENT :

1.1.1. INTRODUCTION :

It has been known for more than a century that cadmium can cause acute poisoning in man. That cadmium could cause chronic poisoning was not established until 35 years ago, when the chronic syndrome after long-term exposure to cadmium oxide dust was described (Friberg, 1949, 1950). Friberg made detailed examinations of cadmium-exposed workers and found that lung damage, mainly emphysema, and renal dysfunction were the major features. Since then, many similar investigations have been performed in several countries and exposure to cadmium compound is now recognised as a serious occupational hazard.

From being a problem only inside the industries, cadmium is now also recognised as a potential hazard to the general population. In Japan, large population groups are exposed to cadmium via food, especially rice. In several areas polluted by cadmium, a high prevalence of proteinuria has been found, and in at least one area, the exposure has been high enough to cause more severe damage - the itai-itai disease.

The metabolic peculiarities of cadmium, i.e., the slow selective accumulation in the kidney and the extremely long biological half-time, make it difficult to extrapolate from the results obtained in short term animal studies. A special effect of cadmium documented in animals is hypertension.

1.1.2. CHEMICAL AND PHYSICAL PROPERTIES :

Cadmium is a chemical element, atomic number 48, atomic weight 112.40, consisting of light stable isotopes of abundance: $^{106}Cd.$, 1.22%; $^{108}Cd.$, 0.88%; $^{110}Cd.$, 12.39%; $^{111}Cd.$, 12.75%; $^{112}Cd.$, 24.07%; $^{113}Cd.$, 12.26%; $^{114}Cd.$, 28.86%; $^{116}Cd.$, 7.58%. Like zinc, and mercury, cadmium is transition metal in group IIb of the periodic table of elements. Cadmium and zinc, however, differ from mercury in that the latter has 14 additional electrons in the fourth orbital, which probably accounts for the high stability of compounds, with mercury-carbon bonds whereas the similar alkyl-cadmium compounds are extremely unstable, reacting rapidly with water and moist air under environmental condition. Unlike mercury, cadmium and zinc show only valence+2 in their compounds; they are also generally similar in reactivity, zinc being the more reactive, and cadmium showing a slightly greater tendency to form covalent bonds especially with sulphur.

The sulphides, CdS (dimorphous, like corresponding zinc and mercury sulphides), and the carbonates, $CdCO_3$, are less soluble than the corresponding zinc compounds, but the hydroxide, $Cd(OH)_2$, is more soluble than $Zn(OH)_2$. Cadmium forms a wide variety of soluble complexes notably with cyanides and amines.

1.1.3. USE PATTERNS OF CADMIUM :

The principal uses of cadmium are as alloys, in plating metals, pigments, as a stabilizing material for polyvinyl plastics, in batteries, fungicides, nuclear control rods, phosphorus, ceramic and others. Data from the U.S. Bureau of Mines (referred to in Fleisher et al., 1974) on U.S. and world production of cadmium are summarized in Table I.

1.1.4. CADMIUM IN AIR :

Data on the concentration of cadmium in air of United States collected by National Air Sampling Network have been summarized by Tabor and Warren (1958), Athanassiadis (1969), and by Schroeder (1970). Schroeder, considering the data collected through 1966 for 58 cities and 29 non-urban areas giving range of concentration (ng Cd/m^3): 2 - 370 for urban areas, 0.4 - 26 for non-urban areas. Friberg et al. (1971)

Table I : World and U.S. production of zinc and cadmium

Year	Metric tons			
	Cd(U.S.)	Cd(world)	Zn(U.S.)	Zn(world)
1971	3,597	15,490	554,000	6,700,000
1970	4,293	16,620	1,053,000	6,680,000
1969	5,736	17,576	1,215,000	6,494,000
1968	4,831	14,674	1,213,000	6,044,000
1967	3,946	13,186	1,116,000	5,876,000
1966	4,745	13,023	1,222,000	5,448,000
1965	4,386	11,676	1,188,000	5,227,000
1964	4,744	12,674	1,131,000	4,872,000
1963	4,491	11,894	1,050,000	4,464,000
1962	5,052	11,903	1,034,000	4,332,000
1961	4,725	11,295	995,000	4,101,000
1960	4,738	11,113	957,000	3,957,000
1955-1959 (avg)	4,487	9,435	1,064,000	3,753,000
1946-1950 (avg)	3,644	4,980	940,000	2,260,000

quote weekly means of 500 ng Cd/m^3 at a distance of 100 m and 200 ng/m^3 at a distance of 400 m from a Japanese smelter, and 160 - 320 ng/m^3 at a distance of 500 m from another Japanese smelter. Weekly means of 600 (maximum 54,000) mg/m^3

at a distance of 100 m and of 300 at a distance of 500 m were recorded in Sweden near a factory using copper-cadmium alloy. Data are not available on the fate of air-borne cadmium and its residence time in the atmosphere. It is presumed to be carried down by rain and snow, but the few determinations made so far are inadequate. The data of Lagerwerff (1971) indicate that near highways, nearly half the cadmium taken up by plants is from air-borne sources.

1.1.5. CADMIUM IN SOIL :

Analysis of uncontaminated soils indicate that normal contents of cadmium are less than 1 ppm perhaps about 0.4 ppm on the average (Fleischer et al., 1974). The data of Lagerwerff and Specht (1971) and Lagerwerff (1971) show clearly the extent of soil contamination near highways. Contamination from smelters and metallurgical plants is even more striking. The contents are significantly higher in the industrial and airport zone than in the residential areas.

1.1.6. CADMIUM IN WATER :

Most fresh waters contain less than 1 ppm Cd. The chemistry of cadmium in surface and ground waters has been reviewed by Hem (1972) who gives calculations of equilibrium solubilities with $Cd(OH)_2$ or $CdCO_3$, showing minimum

solubility at pH 9.0 - 10.0. Duram et al. (1971) found less than 1 ppb Cd. in 54% of the 727 surface water analysed, which had been filtered through 0.54 μ opening. Kopp and Kroner (1968) reported finding no cadmium in 97.5% of the 1577 surface waters analysed. Friberg et al. (1971) point out the need to determine the cadmium content of the suspended matter; they report that 500 m downstream from cadmium emitting factory in Sweden, the water contained 4 ppb Cd. the mud 80 ppm Cd (dry weight).

A study by Perhac (1972) who made ultracentrifuge separation of coarse and colloidal particulate matter from four samples in a mineralized area showed that the particulate matter had concentration of cadmium some thousands of time that dissolved in the waters; yet that 85-96% of the total cadmium was present as dissolved material. These data indicate that cadmium is precipitated on stream sediments under some condition, the extent of precipitation depends upon pH, degree of complexing and many other factors. Abdullah and Royle (1972) report that "clean" streams in Wales contain 0.41 μg Cd/l, and streams affected by old mining activities contained 1.1 - 3.4 μg/l.

The surface water that contains more than few ppb Cd near urban areas have almost certainly been contaminated by industrial wastes from metallurgical plants, plating works,

or plants manufacturing cadmium pigments, cadmium stabilized plastics or nickel-cadmium batteries or by effluent from sewage treatment. Among the few well documented studies on industrial contamination of water supplies by cadmium are those of Lieber and Welsch (1954), who traced the spread of cadmium from a plating plant over an area up to 0.8 mile long and 0.2 mile wide, finding ground water containing upto 3200 ppb Cd.

1.2. NORMAL HUMAN INTAKE OF CADMIUM FROM ENVIRONMENTAL SOURCES :

The principal source of cadmium would appear to be from food rather than air or water (This may not be true in smokers). Cadmium occurs in small amounts in all foods used by man or animals although little is known about its chemical form or binding.

Table II represents some data by Schroeder et al.(1967). Friberg et al. (1971) have reviewed data on cadmium in food in various countries and there seems to be general agreement that food averages about 0.05 ppm Cd (net weight) with of course wide variation depending on the source.

Table II : Cadmium in various classes of foodstuffs

Class of Food	Mean concentration ppm (wet weight)
Seafood	0.79 (oysters excluded)
"	0.33 (oysters and anchovies excluded)
Meats	0.88
Dairy Products	0.27
Cereals and grains	0.19
Vegetables (legumes)	0.03
Vegetables (tubers)	0.07
Vegetables (leafy)	0.13
Oils and fats	0.83
Nuts	0.05
Fruits	0.04
Milk	0.025 (median)

There have been relatively few comprehensive studies of the total human intake via foods, but the available data would put the average of 50 µg/day or less with considerable variation. The best estimates of the U.S. food intake are by Schroeder and Balassa (1961), Murthy et al. (1971) and Duggan and Lipscomb (1969) (Table III).

Table III : Intake of cadmium from food in U.S.

Intake, ug/day (mean or range)	Analytical method	Source of food	Reference
4-6	Dithizone	Random selection	Schroeder and Balassa (1961)
27-64*	Atomic absorption	Institutional diets of children	Murthy et al. (1971)
26		"Market Basket" survey	Duggan and Lipscomb (1969)

* Range of average values.

Further evidence of the approximate correctness of the figure of 50 µg/day is provided by data on daily fecal excretion of cadmium in the general population. Thus Tsuchiya (1969) reported that daily fecal excretion in four non-occupationally exposed men was 57 µg.

The study of water supplies would indicate that except for unusual instances of contamination, the intake via water is probably negligible (Kopp, 1970). The average intake from drinking water is about 1 or 2 µg/day. Little seems to be known as to the contribution of particulate content of water to the total cadmium content.

Airborne particulates or aerosols provide an additional source of Cd to the body. A large amount of data from 35

stations gave an average airborne Cd concentration of 0.002 $\mu g/m^3$ (Fleisher et al., 1974). However, work by Lewis et al. (1972a) on cigarette smoking as a source of cadmium suggests that this may be an important item. Autopsies were performed on 172 adults, including 45 male smokers whose approximate cigarette consumption was known, and 23 non-smoking males. The mean age at death for each group was 60 years. Cadmium levels in lungs, liver and kidney were determined by using atomic absorption. The estimated body burden of cadmium in non-smokers averaged 6.63 mg and was double the amount, 15.8 mg in the smokers. Lewis et al. (1972b) estimate that their data point to a non-smoker retention of 1 µg or less per day, compared with about 2.5 µg/day for smokers. Estimate of total body burden from the data would give non-smokers about 12 mg and smokers about 30 mg at age 60. Szadkowski (1969) reported that about 1.4 µg Cd was found in a cigarette and estimated that 0.1 µg Cd per cigarette would be in particulate phase, 0.03 µg Cd in the gaseous phase. About 0.1 - 0.13 µg might be inhaled per cigarette. The respiratory intake from two packs per day would be about 4 - 6 µg or 10 - 20 times the intake from the reported levels in the air of lower Manhattan.

In summary, it can be stated that intake for men under ordinary circumstances is principally from food, and most estimates would put this at about 20-50 µg/day. Due to poor absorption from intestinal tract it is possible that only about 2 µg/day or less actually is assimilated. The intake from drinking water is presumably 1 or 2 µg/day on the average due to poor absorption, probably only about 0.1 µg/day is assimilated. The intake from ambient air is also probably very low. The daily assimilation of cadmium from ambient air would be approximately 0.02 µg.

From the non-smoking, non-industrially exposed U.S. adult, the likely daily assimilation can be summarized as follows: From air 0.02 µg, from food 2.0 µg, from water 0.1 µg for a total of 2.12 µg. This value approximates the reported daily excretion of cadmium, suggesting that adults in general population are approximately in cadmium balance. This conclusion may be unwarranted because available data on urinary excretion of cadmium do not distinguish between smokers and non-smokers, whereas the estimate of daily assimilation for smokers and non-smokers combined, is probably appreciably higher than 2.12 µg and therefore, probably higher than urinary excretion or perhaps even higher than urinary and fecal excretion combined.

1.3. METABOLISM OF CADMIUM :

Although there is little doubt that man and other mammals absorb cadmium through the lung and mouth, there have been very few studies designed to estimate quantitative human uptake from the environment.

Tipton and Stewart (1970) described a balance study in three normal subjects over periods from 140 to 347 days. Using atomic absorption methods, the amount of cadmium and a number of other metals were determined in the diet, and the amount excreted in the urine and faeces were measured.

The subjects were reported to have ingested an average of 170 µg Cd/day from the diet and to have excreted about 42 µg/day in the faeces and 94 µg/day in the urine. Friberg et al. (1971) point out that these results indicated an absorption of 75% of the intake much higher than the percentage reported by others. The urinary excretion also seemed very high and it seems possible that analytical errors resulted from sodium chloride interference (Friberg et al., 1971).

Bostrom and Wester (1969) studied the intake in a human for short period of 5 days. They found that cadmium intake levels of 12 µg/day in each of the two periods and fecal excretion of about 5 µg/day. The short duration of

the study would make interpretation of absorption uncertain. Rahola et al. (1972) reported on work in which five human subjects were given orally $^{115}Cd(NO_3)_2$ mixed with kidney suspension. The dose used was 100 µg as Cd, containing about 5 ug Ci ^{115m} Cd and body retention was studied by whole body counting, urine and fecal excretion were also followed. About 6% of the dose appeared to have been retained at two weeks. The subsequent rate of decrease in the amount retained in the body was extremely slow. The half-time for body retention was in excess of 100 days. Therefore, the total absorption of oral dose of cadmium probably was not much greater than the 6% found to be retained after two weeks.

Various animal studies also suggest that the absorption of cadmium from the gastrointestinal tract is poor. For example, Decker et al. (1957) found 2.6% of a single oral dose of ^{115}Cd in the liver and kidneys of rats three days later. At 7 and 15 days after administration, 2% of the dose was present in these organs. Thus, at least 2.6% of the dose has been absorbed, but probably not much more than that, in view of the fact that little or no radioactivity could be detected in muscle, lung, bone, spleen and urine. Decker et al. (1958) measured cadmium levels in the kidney and liver. These contained about 0.3 - 0.5% of the dose of

cadmium ingested in one year. Since 50-75% of the body burden will be in these organs it is clear that overall retention probably was only 1% or less of the dose. Lucis et al. (1969) studied the metabolism of ^{109}Cd administration to rats orally. The results of this experiment indicated low excretion rate and storage mainly in liver and kidney. Friberg et al. (1971) have also reported that monkeys retained only about 3% of an ingested dose 10 days after ingestion. Another method of estimating absorption in humans is to determine body burden at autopsy and estimate total intake over a half-time. Though subject to many errors, such calculations are compatible with an absorption rate of about 3-8% (Friberg et al., 1971).

Although it is quite evident for its acute and chronic effect in occupational studies that cadmium is absorbed from the human respiratory tract, the precise degree and condition governing pulmonary deposition clearance, and absorption are unknown. Perhaps, the earliest indication of respiratory absorption came from a report by Stephens (1920). A 67 year old man, who had worked in a zinc smelter for many years, was alleged to have had lead poisoning but his symptoms were atypical. At autopsy no lead was found in the liver but 120 ppm Cd and similar amounts of zinc were present. Eight other similar cases were noted. Subsequent studies of autopsy material from persons with occupational

exposure by Friberg (1950), Bonnell (1955), Smith et al. (1960) and Kazantizis et al. (1963a) showed both liver and kidney to have higher concentration than normal. Lung concentrations were also higher, especially when the exposure was to insoluble pigments (Kazantizis et al., 1963a). Increased blood and urine levels have also been noted in exposed workers (Friberg et al., 1971).

There is strong evidence that cigarette smoking contributes substantially to the cadmium body burden of smokers (Lewis et al., 1972a, 1972b). Rough calculations suggest that the retention of cadmium inhaled in cigarette smoking is substantial. The following calculations lead to such a conclusion.

The body burden of cadmium due to cigarette smoking 0.36 mg/pack-yr (where 1 pack-yr denotes one pack smoked per day for one year) is estimated from the data of Lewis et al. (1972a, 1972b). On estimating that sum of cadmium in liver, kidney and lungs attributable to cigarette smoking is 25 mg/100 pack-yr minus 7 mg from other sources or 18 mg, the total body burden is assumed to be twice the burden in these organs, yielding (18 mg x 2)/100 pack-yr or 360 µg/pack-yr. The finding (Menden et al., 1972) that approximately 2 ug Cd is inhaled per pack leads to a finding that 1 pack-yr provides 730 µg Cd inhaled (2 x 365 = one pack smoked

per day for one year). From these data the percentage of Cd may be estimated.

$$\% \text{ retention} = (360/730) \times 100 = 49\%.$$

In industry, the major hazard from cadmium is from inhaling cadmium oxide fumes. These are probably particles having mass median diameter of 0.5 u or less. Cadmium oxide also is classed as being very poorly soluble. Using the International Commission on Radiological Protection (ICRP) lung model (1966), one would expect roughly 40% deposition and less than 10% retention of inhaled fume particles, mainly because of predicted retrograde movements of particles to the pharynx with ultimate swallowing. There is no experimental confirmation reported for the specific case of cadmium oxide. Thus, the fate of inhaled cadmium is poorly known. In any event, ambient air is only a minor source of cadmium intake for the general population, cigarette smokers excepted.

Estimates of cadmium retention in animals have been made. Prodan (1932) studied the accumulation of cadmium in cats exposed to the fume, oxide and sulphide for short periods. The largest percentages were found in the lungs, liver and kidney. Prodan estimated the retention to vary from about 17 to 30%. In the case of sulphide exposure, virtually all the cadmium was found in the lung. Friberg

(1950) exposed rabbits to a mixture of cadmium and iron oxide dust and found the principal amounts in the lung, liver and kidney and an estimated pulmonary absorption of about 30%.

Little attention has been given to skin absorption. Skog and Wahlberg (1964) applied a $^{115}CdCl_2$ isotope to guinea pig skin and found 1.8% absorbed in 5 hrs. It seems likely that skin absorption is relatively insignificant.

Cadmium is absorbed well from the injection site and again seems to be stored mainly in liver and kidney (Friberg et al., 1971).

The exact mechanism by which cadmium is transported through intestinal mucosa is unknown although there has been some speculation that it may involve mechanism similar to those of copper and iron transport. There now is substantial evidence that once it reaches the liver, its presence stimulates the formation of an unusual protein, of low molecular weight discovered by Margoshes and Vallee (1957) and named metallothionein. It was originally found in equine kidney but now is known to be present in the liver and kidney of many mammalian species. It usually contains about equal molar concentration of cadmium and zinc and has a molecular weight of about 10,500. It may contain as much as 5.9% Cd and also has the ability to bind mercury.

The work by Nordberg et al. (1971) has resulted in the finding of 60% of ^{109}Cd related to a similar protein in mouse blood after repeated subcutaneous injections over a 6-month period. The protein was attached to the red cells, but clearly separable from hemoglobin. The plasma levels were too low to allow separation and identification of the binding protein. Some portion of plasma binding was thought to be on a low molecular weight protein. This fraction may be filtered in the glomerulus and thus perhaps represents the pathways for tubular reabsorption, storage, and urinary excretion.

The excretion of cadmium in the urine has been approximately 1-2 µg/day in the general adult population (Imbus et al., 1963; Lehnert et al., 1969). Little is known concerning the relative importance of urinary and fecal excretion in man. The only study that provides any information indicates that 30 days following oral dose of ^{115}Cd, the rate of fecal excretion was approximately half the rate of urinary excretion (Rahola et al., 1972). There also is some evidence for excretion through the intestinal tract into the feces following parenteral injection in rats (Decker et al., 1957).

The levels of cadmium in blood and certain organs in the general population appear to be fairly well established and reviewed in detail (Friberg et al., 1971; Fassett, 1972b).

Some of the values for blood, liver, and kidney are given in Table IV.

Table IV : Average cadmium levels in blood, liver, and kidney of normal humans

Blood Cd µg/100 ml	Liver Cd, ppm wet	Kidney Cd, ppm wet	Reference
1.8	-	-	Kubota et al. (1968)
0.8	-	-	Imbus et al. (1963)
-	2	11	Curry and Knott (1970)
-	3	50	Schroeder and Balassa (1961)
0.5	2	22	Friberg et al.(1971)

The correlation between the concentration in blood and other organs has not been established for man. Thus, blood cadmium has not yet been shown to be a reliable index of exposure. Concentration in other organs are considerably lower than in either liver or kidney. The body burden of Cd in an adult is estimated to be about 30 mg. The newborne is said to contain only about 1 µg of Cd, so there is a gradual increase with age. Of the total body burden, 50-75% will be in the liver and kidneys, about 1/3 (one-third) of it in the kidney. Normally the kidney concentrations are 5-20 times those of liver. From the scattered data collected

so far, it would appear that the estimated body burden in human varies with age from virtually zero in the newborn to a maximum of 10-18 mg in adults (Friberg et al., 1971). Autopsy data suggest that this maximum is attained in middle life. There is some evidence that the kidney levels may decrease slightly after that. Friberg et al. (1971) have speculated that there may be some loss from the kidney in older age groups but an alternate possible explanation is proposed by Hammer et al. (1972), suggesting that the present older generation may have had lower exposures during their lifetime than the current younger generation. The fact that cadmium accumulates to a large extent in liver and kidney is better established than the range of total body burden and its variation with age.

The concentration of cadmium in the kidneys is of special interest. Industrial experience suggests that the kidneys are especially sensitive to the toxic effects of cadmium. The threshold concentration of cadmium in kidney above which renal damage is likely to occur is estimated to be 200 ppm in outer cortex (Friberg et al., 1971). This is approximately four times the concentration reported for adults in the general population in one study (Schroeder and Balassa, 1961) and about six times the concentration reported for the category of moderate smokers of advanced

age in another study (Lewis et al., 1972a). The margin of difference might be considerably smaller for moderate smokers in middle age, since the concentration of cadmium is known to fall in advanced age.

1.4. TOXIC EFFECTS ON MAN AND ANIMALS :

There are a number of well documented studies of acute and chronic effects of cadmium in man. Animal studies indicate a variety of toxic effects, the significance of which has not yet been demonstrated for man.

1.4.1. EFFECTS IN MAN :

The effects in man can be classified as follows: acute oral, chronic oral, acute inhalation, and chronic inhalation effects.

The acute oral effects are similar to those of zinc, and there have been many well documented epidemics of acute gastroenteritis from the ingestion of acid-type foods that have been stored in cadmium-lined containers (Fairhall, 1957). The dose causing the symptoms has been estimated (Fleisher et al., 1974) to be as low as 15-30 mg. The symptoms come on almost instantly after ingestion of the contaminated food, and the acute nausea and vomiting may in some cases be followed by a severe gastroenteritis. The

actual oral lethal dose in man has not been established, but estimates have been made that it is probably in the neighbourhood of several hundred milligrams (Fleisher et al., 1974).

As far as is known at present, no chronic toxic effects (except for a Japanese incident) have ever been reported from the oral ingestion of cadmium by human subjects. It is of course possible that some of the chronic toxic effects by inhalation may have been due to swallowing of the material being cleared from the lung.

The second important acute toxic effect in man is that of acute pulmonary edema induced by inhalation of metallic fumes or cadmium oxide dust (Friberg et al., 1971; Fassett, 1972a). It has been estimated that fatalities have occurred from a 5 hr exposure at about 8 mg/m^3 although in one instance recovery was reported after exposure to 11 mg/m^3 for 2 hr. In one brief account of a few intermittent exposures to cadmium fumes during silver soldering, a non-fatal acute pneumonitis was noted at concentrations estimated to have varied between 0.5 and 2.5 mg/m^3 over a 3-day period (Hygienic Guides Committee, 1962).

The acute pulmonary changes seen in man have been reproduced in experimental animals (Nordberg et al., 1971). Evidence does not indicate that acute renal injury is a part

of the picture of acute inhalation toxicity with cadmium, nor is it known whether repeated acute exposures would subsequently result in chronic renal or lung injury.

The chronic exposure to cadmium through the respiratory tract produces a number of toxic effects, the most important of which is the chronic emphysema first described in the classical report by Friberg (1950). Along with the emphysema, a peculiar renal disturbance has been noted, with excretion of relatively low molecular weight proteins in the urine together with some increase in amino acids and at times glucose and calcium. These findings have now been confirmed in a number of other publications and have been reviewed in detail by Friberg et al. (1971). Among the more important of these studies are those by Princi (1947), Bonnell (1955), Smith et al. (1960, 1961), Kazantzis et al. (1963 a), and Piscator (1962, 1966).

It is apparent that chronic cadmium emphysema appears only after a period of exposure averaging about 20 years. It was not possible in most of these studies to arrive at the exact exposure levels since in some cases these may have occurred many years previously. However, from Friberg's initial study it would appear that exposure to cadmium oxide dust at levels of about 3-15 mg/m^3 may have been principally responsible. In a study (Teculescu and Stanescu, 1970) of

11 subjects (eight of whom were smokers) exposed to cadmium oxide in the course of extracting cadmium from master alloys, no physiologic disturbances compatible with emphysema were noticed in the group. However, the length of exposure varied between 7 and 11 years, and the air concentrations were lower than those described in Friberg's original work (1.21 - 2.70 mg CdO/m^3). Elevated excretions of cadmium in the urine were noted, varying from about 3 to 65 µg/24 hr. Although this study included an extremely thorough analysis of all aspects of respiratory function, it is unfortunate that no renal function or urinary protein levels were determined.

Before Friberg began his studies, it had not been shown that cadmium could cause chronic renal dysfunction. The tests used for detection of proteinuria at that time were the boiling test, Esbach's test, or the nitric acid test, which generally were negative in examinations, of urine from cadmium workers. Friberg found, however, by using trichloroacetic acid, that cadmium exposed workers had a high prevalence of proteinuria compared to controls. Further studies showed that this proteinuria was not of the classical type seen in chronic nephritis or nephrosis. The proteins had a different electrophoretic pattern and had lower molecular weight than albumin. A decreased ability to concentrate the urine and a reduction in inulin clearance

were other signs of renal dysfunction (Friberg, 1949, 1950). At that time, it was not possible to further characterize and identify this type of proteinuria, but after Butler and Flynn (1958) had shown that some diseases with congenital or acquired renal tubular dysfunction had a typical urine protein pattern on electrophoresis, it could be demonstrated that the proteinuria in chronic cadmium poisoning was of the same type (Piscator, 1962, 1966).

This tubular proteinuria is characterized by the excretion of low molecular weight serum proteins in the urine, caused by a decrease in reabsorption of proteins from the glomerular filtrate. Improved methods for the determination of total urine protein made it possible to show that the excretion of protein was related to exposure time, as shown in Table V. This low molecular weight proteinuria is regarded as the first sign of an effect of cadmium on renal function, and by using immunological methods for determination of specific proteins in combination with electrophoresis, it is now possible to detect small increases in excretion of such proteins, e.g., β_2-microglobulin. The other signs mentioned earlier in this section, e.g., glucosuria and aminoaciduria, generally appear later than the proteinuria. The renal dysfunction will also cause an increase in the urinary excretion of cadmium (Friberg et al., 1974).

Table V : Urinary protein excretion in cadmium-exposed workers in relation to exposure time

Exposure time (years)	N	Urinary protein excretion (mg/24 hr)	
		Mean	Range
0	15	60	25-110
1-5	4	100	50-170
6-10	4	210	70-570
11-15	5	300	70-770
16-20	9	575	165-1300
21-25	6	460	160-1050
26-30	6	790	210-2600
31-	6	955	370-1800

From Piscator (1962).

In investigations on population groups exposed to cadmium via rice in Japan, it has been shown that the proteinuria is of the same type as seen in cadmium-exposed workers and that cadmium excretion is considerably higher than in people from nonpolluted areas (Friberg et al., 1974).

The critical concentration of cadmium in the renal cortex, i.e., the concentration at which sensitive individuals may get tubular dysfunction, has been estimated to be 200 µg/gm weight (Friberg et al., 1974).

This estimate was based on data obtained from autopsies on former cadmium workers and agrees well with findings in animal studies.

There may also have been some increase in incidence of renal stones in those with long exposure. Ahlmark (1961) also reported a high incidence of renal stones with long exposure to cadmium dust. Nicaud et al. (1942) described X-ray changes characteristic of pseudo fractures in certain workers exposed to cadmium oxide dust. Findings of this type were not noted in later studies made by Friberg. Kennedy (1966) studied serum calcium levels in rabbits given repeated injections of cadmium. There was a very slight fall in serum calcium which was interpreted as being due to possibly increased renal excretion. Another finding in the work of Kazantzis (1956) is that there was evidence in some individuals of increased output of calcium in the urine.

The form in which cadmium is excreted in the urine is not known although it is possible that it may be in the form of a metallothionein complex.

Slight anaemia has been noted in some subjects and it is thought possible that this might be related in some way to interference with zinc, copper, or iron metabolism. At present no explanation is available for the appearance of anaemia. Anosmia has been reported in a group of alkaline

battery workers who were exposed to both cadmium and nickel dust (Adams and Crabtree, 1966).

In 1955, a somewhat unique disease was described in the vicinity of a mine in Toyama Prefecture, Japan. The disease was epidemic among elderly women who had borne many children (average of 6). The outstanding features of the disease were lumbar pain and myalgia, spontaneous fractures with skeletal deformation. Pain was readily elicited from pressure applied to bones. Extensive epidemiological studies were instituted after it was demonstrated that the water, rice, and fish in the endemic area were found to contain high concentrations of cadmium and other metals, probably due to contamination of the local river by the effluent from a zinc-lead-cadmium smelter. These studies continue and have been extended to include other areas in Japan where similar mining operations exist. The results of these investigations have been summarized by Friberg et al. (1971).

The evidence available to date strongly indicates that this syndrome, termed itai-itai, is due to long-term cadmium exposure. It is the first likely instance of cadmium poisoning in man due to general environmental contamination. The characteristic skeletal changes found in the older women are not usually observed in industrial cadmium poisoning. They are ascribed to the interplay of cadmium

exposure and certain other factors not usually encountered in industrially exposed groups, such as old age, low nutritional status, and multiparous motherhood. The neuromuscular signs and skeletal defects described in itai-itai, however, have also been observed in a series of cases of cadmium poisoning in France during World War II. Four women and two men were affected. All had been exposed for at least 8 years (Nicaud et al., 1942). Other cases of this type in industrial cadmium poisoning also have been reported (Bonnell, 1955; Gervais and Delpech, 1963; Adams et al., 1969). Thus, the musculoskeletal features of itai-itai are far from unique as manifestations of excessive cadmium exposure.

Itai-itai is not solely a musculoskeletal disease. It is accompanied by the more classical renal effects of cadmium seen in industrial poisoning. Proteinuria was always found in clinical cases of itai-itai. Glucosuria and aminoaciduria also usually were present (Friberg, 1971). Further, the incidence of proteinuria and glucosuria was much higher among older women and men in the endemic area than elsewhere in Toyama Prefecture. The urinary excretion of cadmium also was three times greater among people in the endemic area than in the nonendemic area of Toyama Prefecture. The prevalence of proteinuria and glucosuria was only somewhat

greater among women than among men in the endemic area. Thus, while the musculoskeletal manifestations of itai-itai are seen almost exclusively among older women, the renal manifestations do not appear to be sex-related.

It has not been possible to develop from the available data a dose-response curve for itai-itai. Data on dietary intake of cadmium in the endemic area have been available only for recent years. The recent daily oral intake has been estimated to be 600 µg, ten times the estimated intake for the general population in Japan (Yamagata, 1970). It seems likely that exposure of that order or higher occurred for at least 20 years and that such long-term exposure may have been necessary to attain the recent incidence of proteinuria among older women in the endemic area. Women born in another area but residing in the endemic area for 20 years or more have an incidence of proteinuria not quite as great as those born and living in the area all their lives (Friberg, 1971). The statistical significance of the difference in the incidence of proteinuria in these two groups has not been reported.

Studies conducted in Japan indicate that excessive exposure to cadmium may be more widespread in that country than had been previously thought (Friberg et al., 1973). Unfortunately, due to faulty experimental design, these

studies have not provided any significant new information in regard to dose-response relationships.

1.4.2. EFFECTS IN ANIMALS :

Experiments on animals have verified that exposure to cadmium causes renal tubular dysfunction (Axelsson and Piscator, 1966; Nordberg, 1972). Autoradiographic studies by Berlin et al. (1964) showed that cadmium accumulated mainly in the proximal part of the proximal tubule. This is the part of the tubule where protein reabsorption occurs. It has also been indicated that cadmium bound to the cadmium-binding protein metallothionein may be filtered through the glomeruli and reabsorbed (Nordberg, 1972). It is conceivable that cadmium originally bound to this protein in the liver will in that way be transported to the renal tubules and that cadmium will accumulate mainly at a site where there is reabsorption of protein. This also means that the first function to be disturbed when there is excessive accumulation of cadmium should be protein reabsorption, which fits well with the experience from investigations on human beings.

It has also been shown (Nordberg, 1972) that in mice exposed to cadmium, the urinary excretion was very low as long as the renal function was normal, whereas there was a sharp rise in cadmium excretion when the tubular proteinuria

appeared. It was then also possible to demonstrate the presence of cadmium in a low molecular weight urine protein fraction, conceivably metallothionein. That means that the decreased reabsorption of proteins also causes a decrease in the reabsorption of metallothionein. This increase in cadmium excretion may eventually lead to a depletion of renal cadmium as seen in itai-itai patients.

Data from animal experiments also indicate that the critical level in renal cortex is around 200 ug/gm wet weight (Axelsson and Piscator, 1966; Nordberg, 1972; Stowe et al., 1972). In calcium-deficient rats, however, the excretion of the low molecular weight enzyme ribonuclease was increased at a cadmium concentration in renal cortex of about 90 µg/gm wet weight, indicating that under certain circumstances the critical level might be below 200 µg/gm (Piscator and Larsson, 1972).

One of the most interesting effects of cadmium in animals studies has been its ability to cause an acute necrosis of the rat testis following either parental injections or relatively large sublethal oral doses. This matter has been reviewed (Friberg et al., 1971; Fassett, 1972b). Studies by Parizek (1957) first established the extraordinary protective effect of zinc treatment against the acute atrophy of the testis resulting from cadmium

injections. This has also been studied by Gunn and his co-workers (1968). Cysteine and selenium also will protect against the cadmium testicular atrophy. By use of ^{109}Cd, it was shown that none of the protective agents actually lowered the amount of cadmium reaching the testis. Anatomical studies have demonstrated (Mason et al., 1964) that in all probability this acute cadmium testicular damage results from a toxic effect on the unique vascular system of the testis.

Because of the well-known effect of cadmium on the testis, a number of investigators have looked at the effect of cadmium on the course of pregnancy and fertility. Most of these studies have been based on cadmium injection. Parizek (1964) showed that there could be fairly rapid destruction of the fetal placenta with only slight effects on the maternal placenta. When selenium salts were simultaneously injected, no placental effects could be produced. A teratogenic effect of cadmium has been demonstrated in the hamster (Ferm and Carpenter, 1967). This effect was inhibited by zinc.

There is no evidence at present that the testicular effects found by injection in experimental animals are to be found in man. Friberg et al. (1971) quote Cvetkova (1970) as having reported somewhat lower weights in male and female

children born to women working in a cadmium accumulator factory with relatively high levels of exposure to cadmium. Obviously many more studies of this sort are necessary before any conclusions can be drawn as to human effects of this type.

Studies of possible carcinogenic effects of cadmium have been primarily in experimental animals. The tumours found mostly have been of the sarcomatous type and have mainly been localized at the site of injection (Heath et al., 1962; Kazantzis, 1963b; Haddow et al., 1964; Heath and Daniel, 1964; Kazantzis and Hanbury, 1966). In two of these studies metastatic tumours occurred in the regional lymph nodes and in the lungs (Kazantzis, 1963b; Kazantzis and Hanbury, 1966). Interstitial cell neoplasms have been shown to develop in the testes of rats, both after direct injection into the testicle and after subcutaneous administration (Gunn et al., 1965; Lucis et al., 1972). Long-term feeding of cadmium also has been shown to increase the incidence of neoplasms in rats but not in mice (Schroeder et al., 1964; Schroeder et al., 1965; Kanisawa and Schroeder, 1969b). Kazantzis (unpublished data referred to in Fleisher et al., 1974) reported that long-term feeding studies in rats nearing completion have not yet revealed any evidence of carcinogenic activity.

Other studies reviewed by Shubik and Hartwell (1951, 1957, 1959) and studies by Decker et al. (1958) and Anwar et al. (1961) did not reveal evidence of cadmium-related cancer formation in animals.

In view of the frequent demonstration of a carcinogenic effect in experimental animals, the implications for a carcinogenic effect in man need to be explored thoroughly. Indeed, some limited epidemiological studies have been conducted in industrially-exposed populations. Potts (1965) reported a high incidence of cancer among 74 men exposed to cadmium for at least 10 years. Three of the eight men who died had prostatic cancer and two had other forms of cancer. The incidence of cancer of the prostate was also reported to be high in another study of industrially exposed workers (four cases versus an expected number of 0.58) (Kipling and Waterhouse, 1957). Malcolm (1972) concluded that cadmium does not cause cancer of the prostate in man in spite of the suggestive evidence.

In view of the relatively high concentration of cadmium in cigarette smoke, its possible role in bronchogenic carcinoma has received some consideration (Friberg, 1971) but a causal role has not yet been demonstrated.

Finally, one of the most interesting aspects of the cadmium toxicity is the discovery by Terhaar et al. (1965)

that pretreatment with very small oral doses of cadmium would protect against the effects of subsequent very large or fatal doses of cadmium chloride. Doses as low as 10 µg/kg given orally 24 hr before a subsequent dose of 100 mg/kg completely protected against the massive testicular atrophy. Others have since found similar protective effects of pretreatment with small doses against a variety of lesions (Gabbiani et al., 1967; Yoshikawa, 1969).

It now seems almost certain that the nature of this remarkable protective effect is due to the induction of metallothionein in the liver by subcutaneous or oral administration as demonstrated by Shaikh and Lucis (1970). The ability of cadmium to induce the formation of such a specific protein has raised a question as to whether this is not a fundamental protective mechanism against toxicity from this and perhaps certain other metals. Although metallothionein has thus far only been isolated from mammalian tissues, Maclean et al. (1972) reported recently on the uptake of $^{109}CdCl_2$ and of $^{65}ZnCl_2$ by various microorganisms. The organisms involved were E. coli, Anacystis nidulans, Chlorella, Crithidia fasciculata, and Chondrus crispus. Under the conditions of this study, these bacteria and algae were all able to extract both cadmium and zinc from the solution. Protein separations indicated that both the zinc and the cadmium were bound to a protein with similar

properties to the metallothionein isolated from mammalian tissues. Whether such proteins represent the form of cadmium in plants is unknown but this seems possible.

However, the apparently nearly universal occurrence of metallothionein or similar proteins containing roughly equal moles of cadmium and zinc and the obvious "evolutionary antiquity" of this type of protein continue to make this an important question. A review (Anonymous, 1968) of cadmium in the metabolism of albumin suggests that perhaps cadmium ions may play an important physiological role in regulating the biosynthesis of albumin and perhaps other proteins including some enzymes.

Since osteomalacia has been found both in exposed workers and in Japanese women exposed to cadmium via rice, special attention has during the last few years been paid to different aspects of cadmium and mineral metabolism. The concentrations of cadmium in bone tissue are low even at high body burdens, and there is little evidence for a direct action of cadmium on bone. In short-term experiments on calcium-deficient rats given cadmium in drinking water (25 µg/gm for 2 months) and long-term experiments (7.5 µg/gm for 13 months), calcium accretion in bone was not decreased in cadmium-exposed animals (Larsson and Piscator, 1971; Piscator and Larsson, 1972). It was found, however, that

cadmium accelerated the osteoporotic process caused by calcium deficiency alone, and both interference with intestinal absorption of calcium and endocrine disturbances might have caused this. In the short-term experiment, it was found that the increase in parathyroid volume, which is caused by calcium deficiency, was inhibited by cadmium. In the long-term experiment, there were signs of slight renal tubular damage already at a cadmium concentration in renal cortex of about 90 µg/gm wet weight, indicating that kidneys of calcium-deficient animals may be more sensitive to cadmium than normal animals.

The bone changes in itai-itai disease have occurred mainly in women with low intakes of calcium and losses due to multiple pregnancies. The exposure to cadmium has caused renal tubular dysfunction, which has caused further losses of minerals. In Swedish factory workers with renal tubular damage but high intake of calcium, bone changes were not seen (Friberg, 1950).

Since the most active form of Vitamin D is produced in the kidney , it is also conceivable that the accumulation of cadmium may interfere with the synthesis of this form, which may further contribute to the disturbances in mineral metabolism.

Flick et al. (1971) reported that cadmium may result in

hemorrhagic lesion in sensory ganglia within the central nervous system. According to Arena (1963), chronic effects of inhaled cadmium include total or partial loss of smell, coughing, laboured breathing, depressed appetite, weight loss, and in many instances a yellow staining of the teeth and generalized irritability. Browning (1961) noted that toxic levels of cadmium by inhalation varied from approximately 1-0 to over 30 $\mu g/m^3$ of air. Fatigue, dental caries, gastro-intestinal upset, pallor, and in 2 out of 5 cases, a low level of blood hemoglobin were also observed. The influence of cadmium on aortic lipid was reported by Schroeder and Balassa (1965). In general, it was found that 5-0 ppm cadmium incorporated into the drinking water of rats of both sexes from weanling unto death resulted in a decrease in circulating cholesterol (more in male than female), an increase in extractable aortic lipid, and an increase in aortic plaque formation.

Recently, Webster and Valois (1981) studied the simple subcutaneous injection of cadmium chloride in saline on postnatal days 1, 8, 15, or 22 on mice. Histopathological examination 24 h after cadmium exposure on day 1 revealed petechial hemorrhages, edema, and cellular pyknosis throughout the immature brain. Treatment on day 8 to 15 produced similar damage particularly edema and pyknosis;

by 22nd day the brain was unaffected by cadmium. Powell (1964) studied the effect of cadmium in various concentrations ranging from 40, 160, 640, and 2560 ppm on calves. Growth rate, feed consumption, water intake and testicular development, decreased progressively as the concentration of cadmium in diet increased. Blood hemoglobin decreased slightly when 40 to 160 ppm cadmium in the diet were consumed, but increased sharply when the higher level of cadmium (640 and 2560 ppm) were fed. Calves fed 64 or 2560 ppm cadmium exhibited unthrifty appearance, rough hair coat, severe body dehydration, dry and scaly skin, loss of hair, mouth lesion, edematous, shrunken and scaly scrotum, sore and enlarged joints, impaired sight, and liver and kidney damage. Feeding 40 to 160 ppm of cadmium from 9 to 20 weeks of age resulted in few clinical symptoms. There was 100% mortality in calves, given 2560 ppm cadmium with death occurring at 2 to 8 weeks, at the level of 640 ppm there was some mortality. When cadmium was removed from the diet, a very sharply affected calf recovered to normal.

Table VI lists some of the more important studies for various species.

Food is generally the most important source of cadmium in the environment, where it occurs in close association with zinc. The average intake from food is probably not more than 60 µg/day. Drinking water and ambient air contribute relatively little to the daily intake under normal circumstances. Airborne cadmium may become significant because of the probability of a higher percentage of respiratory absorption than of oral absorption. However, to date there is no evidence to suggest that ambient air concentrations of cadmium are presenting a hazard. This may not be true, however, if one considers the rather striking potential increase from cigarette smoking. It is evident that in future epidemiologic studies will have to be much more carefully planned to rule out incidental effects such as smoking, and they should take into account all the known facts regarding cadmium metabolism and toxicity.

An estimate of the safe daily intake in the diet is at present very difficult because of a lack of appropriate information. This is especially true with regard to the nature of the chemical bond of cadmium in plants and meat and other foods. Predictions of toxic effects are also rendered difficult by the nearly universal occurrence of very much higher levels of zinc in combination with cadmium and the protective effect of zinc against cadmium toxicity.

The question of a buildup in the food chain similar to that seen with mercury compounds has been raised but there are reasons to believe this is unlikely. Fassett (1972b) has pointed out there are fundamental differences in the electronic structure of cadmium and that of mercury. Mercury is able to form extremely stable carbon-mercury bonds, probably because of the presence of an additional 32 electrons, as compared to cadmium (Sidgwick, 1950). Although alkylcadmium compounds are known, they are extremely unstable and very unlikely to be formed or exist for very long in the natural environment. Furthermore, the methyl-mercury that is build up in the food chain is virtually completely absorbed from the intestinal tract, whereas the cadmium-protein complexes and other forms of cadmium are absorbed only to a very limited extent. In the unlikely event that an individual were to derive his sole source of protein from liver, kidney, or oysters, it is conceivable that the dietary cadmium intake might become hazardous. Until more is known about the absorption of such compounds, however, speculation is very tenuous. The accidental contamination of water supplies may present a very real hazard as well as that of the ambient air. The new data on smoking deserve a thorough investigation.

The role of metallothionein and its induction by small increases in cadmium intake is imperfectly understood.

Cadmium bound as metallothionein may be biologically unavailable. If this is indeed the case, evaluation of the toxicological significance of cadmium levels in tissues (particularly the kidney) will require that the implications of binding as metallothionein be thoroughly explored.

There is no evidence to suggest that the general population is in imminent danger of excessive cadmium exposure. However, many people (particularly heavy cigarette smokers) probably have approximately one sixth to one fourth of the minimally toxic concentration of cadmium in their kidneys. This margin of safety may not be adequate since the minimally toxic concentration is estimated on the basis of very limited data.

1.5. CADMIUM-INDUCED HYPERTENSION :

Some thirty years ago, when the drug treatment of hypertension was in its infancy, Henry Schroeder and Mitchell Perry, Jr., became interested in a group of antihypertensive compounds which act apparently by reducing peripheral resistance (Perry, 1976). These compounds were of a diverse group, and their only obvious common characteristic was their ability to bind transition trace metals. Tipton and Cook (1963) first emphasized the surprisingly high levels of cadmium which were found virtually in all adult human kidneys.

Comparing the concept of metal binding anti-hypertensive agents with the observed accumulation of renal cadmium, led to the speculation regarding a possible role of cadmium in the pathogenesis of human essential hypertension. Since then both Schroeder's laboratories in Brattleboro, Vermont and the laboratories of the Hypertension Division at Washington University in St. Louis have devoted considerable effort to studying the effects of cadmium on the cardio-vascular system and blood pressure.

1.5.1. ANIMAL DATA :

The induction of hypertension in animals by feeding cadmium was first reported by Schroeder and Vinton (1962). Certainly parental cadmium could induce acute transient hypertension in animals (Schroeder et al., 1966; Perry and Erlanger, 1971b). During the next twelve years, Schroeder extended his initial observation in numerous reports (Schroeder, 1964; Schroeder and Buckman, 1967; Schroeder et al., 1968a, 1968b; 1970; Kanisawa and Schroeder, 1969a. Perry and Erlanger (1971a, 1974a) confirmed that chronically fed cadmium could raise systolic pressure. Schroeder et al. have reported very marked hypertension whereas Perry et al. could induce a mild elevation in blood pressure. A third laboratory has reported

mild hypertension following cadmium feeding (Petering et al., 1979). On the other hand, Friberg et al. (1971) have indicated their inability to induce hypertension with fed cadmium.

Schroeder's first experiment began in 1960; after 6 months, the blood pressure of the control and cadmium fed groups were 106 and 218 mm Hg respectively (Schroeder and Vinton, 1962). Two comments can be made regarding significant limitation of these experiments. First, the cited averages involved groups of only eight rats each, and for the cadmium fed group the corresponding standard deviation was large (40 mm Hg). Second, the marked liability of the cadmium-induced hypertension was emphasized and multiple pressures were obviously obtained on each animal; however, nothing is mentioned about the schedule and rules according to which these multiple pressures were taken, although such things could drastically alter the final average pressure.

Schroeder's second report (1964) considers the pressures obtained after 18, 24, and 30 months of cadmium exposure of these animals. Again the marked hypertension in the cadmium exposed rats in comparison to the control rats is evident all three times.

Schroeder's third experiment (Schroeder and Buckman, 1967) consisted of two groups of cadmium exposed rats -

hypertensive and normotensive group, for which the average systolic pressures were 169 and 98 mm Hg. respectively. Since there were nine rats in the former group and thirteen rats in the latter, the average systolic pressure for all twentytwo rats exposed to 5 ppm cadmium was 127 mm Hg. This value is strikingly similar to the average pressure obtained by Perry and Erlanger (1974a).

Schroeder's next report (Schroeder et al., 1968a) showed significant hypertension since the fraction ofthe rats with systolic pressures was greater than 140 mm Hg. After 12 months of cadmium ingestion, 38% of Schroeder's animal had systolic pressure greater than 140 mm Hg. Perry and Erlanger (1974a) in their first experiment used 32 control and 32 cadmium fed rats. Schroeder's experimental condition including acrylic plastic cages, were used. During the first 18 months of the experiment, the average systolic pressure of the cadmium exposed rats remained relatively constant. Although the differences in pressure were small, cadmium feeding was associated with significant increase ($P < 0.001$). By 24 months, the average pressure of the cadmium fed and control groups seemed to have increased, and the difference between them increased to 29 mm Hg (Perry and Erlanger, 1974a).

The second experiment by Perry and Erlanger (1974a)

was conducted simultaneously. It involved considerably more animals and a wide range of cadmium exposures. The rats were housed in stainless rather than plastic cages. During the first 18 months, the average pressures for both the control and the 5 ppm cadmium groups were somewhat above the comparable averages for rats in plastic cages. More important, however, the difference between the control and the cadmium-fed groups were significant with $P < 0.001$; for rats in steel cages as it had been for rats in plastic cages. The 1 ppm group resembled the control group during the first 6 months, the pressures of all three groups had increased but the cadmium fed animals continued to have significantly higher pressure.

Perry et al. (1977, 1979) used groups of 16 female long-Evans rats which received 0, 1, 2.5, 5, 10, 25 and 50 mg/litre drinking water (parts per million, ppm), from the time they were weaned, until they were 30 months old. Systolic pressure was measured indirectly in triplicate at 6-month intervals. A modest but statistically significant elevation in systolic pressure was observed continuously at 6, 12, 18, 24 and 30 months in those groups of rats that had a life long exposure to drinking water containing 2.5 and 5 ppm cadmium. Further significant elevations were observed sometimes in those group of rats receiving water containing 1, 10, 25 and 50 ppm cadmium.

After 6 months under their (Perry et al., 1977) experimental condition which included the standard diet, the average systolic pressure of 64 control rats was 112 mm Hg and the average systolic pressure of the 80 rats fed 2.5-25 ppm cadmium was 125 mm Hg. Although the difference was only 13 mm Hg it was statistically highly significant with $P < 0.001$. After 12 months of cadmium exposure, the rats receiving 10 and 25 ppm cadmium no longer had significant increase in systolic pressure. However, those receiving 1 ppm cadmium had developed hypertension. Thus at 12 and 18 months the survivors among the 64 rats in the group fed 1, 2.5, and 5 ppm cadmium- had systolic pressures that averaged 17 and 16 mm Hg respectively, higher than those of the survivors among the 64 control rats with $P < 0.001$. At 24 months there were 37 survivors in three cadmium-fed groups and 41 surviving controls. The difference in mean systolic pressure between cadmium-fed and the control rats was 22 mm Hg with $P < 0.005$.

Combining the data on systolic pressure level and renal cadmium concentration suggested that under the experimental condition used here (Perry et al., 1977) concentrations of 5-50 µg of cadmium/g of kidney were generally associated with elevated pressures, whereas higher and lower concentrations generally were not.

In a recent study, Kopp et al. (1982) reported the effect of low doses of cadmium in drinking water on blood pressure. A parabolic curve with a maximum (20% increase) at 0.5 ppm Cd was obtained when the blood pressure change was plotted against the logarithm of cadmium concentration in drinking water. This represents a cadmium intake of about 10 µg/day in rats or about 25 times the average North American intake on a per kilogram basis. Further, the effects of cadmium exposure on heart function, and tissue metabolism (high-energy phosphate level) were investigated in rats receiving 1 and 5 ppm cadmium in drinking water (Kopp et al., 1982). The dose dependent changes were consistent with the change in blood pressure with dose, in that cadmium at 1 ppm had more pronounced effect than cadmium at 5 ppm. Conversely, liver metabolism, as measured by changes in high energy phosphate compounds, was little altered by the dose used here.

Occasionally, however, there seemed to be a sub-population of responders in which cadmium induced marked hypertension (Perry et al., 1979). After 18 months, one-sixth of the rats exposed to 0.1, 0.25, and 0.5 ppm cadmium had systolic pressures which exceeded the control mean by 50 mm Hg. In contrast, none of the litter-mate controls or 5 ppm cadmium rats had this degree of hypertension.

Petering et al. (1979) used Sprague-Dawley weanling rats housed in stainless steel cages and provided at libitum Purina rat Chow and deionized distilled water containing 0, 4.3, 8.6, or 17.2 ppm cadmium. Systolic pressure was measured indirectly in anaesthetized animals four times between twenty-ninth and thirty-ninth week of exposure. After 36 weeks, when the maximum effect and stability had been achieved the mean systolic pressure for twelve control males was 117 mm Hg while for 8 males receiving 17.2 ppm cadmium, it was 132 mm Hg. The difference was significant ($P < 0.05$). Boscolo et al.(1981) exposed 24 adult male Sprague-Dawley rats to Cd (as acetate) in deionized drinking water at concentrations of 0, 10, and 20 µg/ml respectively for 160 days. The cardiovascular determinations were made in each rat anaesthetized with sodium pentobarbitone. The arterial blood pressure was continuously monitored by means of Statham pressure transducer connected to a Beckman R.M. Dynograph recorder. Arterial blood pressure was significantly elevated in the Cd exposed rats. The mean values for the diastolic pressure increased from 106 ± 7 mm Hg ($X \pm$ SD) in the control group to 126 ± 14 and 135 ± 22 in the group exposed to 10 and 20 ppm Cd respectively. Similarly systolic blood pressure increased proportionately.

Fadloun and Leach (1980) used male Sprague-Dawley

rats (200-300 g) to study the effect of cadmium on sympathetic nervous control of vascular and nonvascular tissue. The blood pressure of rats treated with Cd (12.5 or 25 ppm) in drinking water for 4 weeks showed marked increase in systolic and diastolic blood pressure when compared to the control which received [illegible] tap water (173/130 mm Hg at 25 ppm Cd^{+} and 130/110 mm Hg control). Revis (1978) could also induce hypertension in rats with cadmium chloride after it was given in drinking water for 60 days (30 or 800 μM).

Two or three negative reports are worth mentioning regarding cadmium and hypertension. Castenfors and Piscator (unpublished data referred to in Friberg et al., 1971) gave Schroeder's usual dose, 5 ppm cadmium in drinking water to Sprague-Dawley rats for 1 year and determined blood pressure by a direct method at a monthly interval; they found average systolic blood pressure of 153 mm Hg for thirteen cadmium-exposed and 151 mm Hg of nine control rats. The average pressures of these control animals were much higher than those found by Schroeder or other groups.

In the second negative report, Lener and Bibr (1970) failed to induce hypertension in Wistar rats given 5 ppm cadmium in deionized drinking water for 16 months, while control animals apparently received water with 1% sodium

chloride, a small amount of cobalt, copper, manganese, molybdenum and zinc. When the blood pressure was measured, there was no significant difference between the control and cadmium treated group. The study by Doyle et al. (1975) showed that 5 µg Cd/ml of drinking water did not produce hypertension after 340 days of treatment.

First report of chronic hypertension after the injection of cadmium was given by Schroeder et al. (1966). Cadmium acetate (2 mg/kg) was administered intraperitoneally to female rats of the Long-Evan's strain. Three weeks after a single injection of cadmium 31% of the rats exhibited elevation of blood pressure. When a second dose of cadmium 1 mg/kg was given to the normotensives, all exhibited hypertension after one week. When cadmium was given intraperitoneally (2 mg/kg) to another group of rats with partial constriction of left renal artery, 71% of the rats showed severe hypertension. Recently, Fadloun and Leach (1981a) developed chronic hypertension in Sprague-Dawley rats by administering cadmium chloride intraperitoneally (0.1, 0.5, and 1 µM) daily for 12 days. In another report (Watkin, 1979), chronic systolic pressure studies were conducted in conscious female Sprague-Dawley rats. Separate groups of 250 g rats received single intraperitoneal injection of 0, 100, 200, 1000 and 2000 ug cadmium acetate. All doses

of cadmium temporarily interfered with normal weight gain in surviving rats. Systolic blood pressure transiently increased by 9-12 mm Hg between 1-6 days after treatment with all Cd doses. The rats which received 100 ug Cd were given 4 additional doses at 14 days' interval. This injection produced similar systolic blood pressure elevation ($P < 0.05$).

Recently, it was shown by Puri and Sur (1983) that cadmium acetate (1 mg/kg,i.p.) for one week can cause severe hypertension in rats of Charles Forster strain. Cadmium injected rats showed blood pressure of 161 ± 8 mm Hg while the control rats had blood pressure of 92 ± 5 mm Hg ($P < 0.01$). Blood pressure remained elevated to statistically significant level ($P < 0.01$) after stopping cadmium for 2 days. Thind et al. (1969, 1970a) produced persistent hypertension in 100% of rabbits by weekly intraperitoneal injection of 2 mg/kg of cadmium acetate for 9 weeks. Further, hypertension was induced in female Mongrel dogs (Thind et al., 1973). Eight adult healthy dogs were given 24 intraperitoneal injection of cadmium acetate 2 mg/kg body weight over a period of nine months. It was found that there was no mortality or significant morbidity or change in the total body weight of the cadmium treated dog. The mean arterial pressure was significantly elevated ($P < 0.05$) in all the dogs.

However, Porter et al. (1974) could not induce chronic hypertension in rats given cadmium intraperitoneally at several dose levels.

Acute pressor response to cadmium in rats was first reported by Schroeder and Perry (1955). It followed the intravenous injection of 50 μg of cadmium ion per 100 gm of body weight. It was further reported that cadmium (0.35 μM) when given intravenously produced initial transient depressor effect in rats prior to its pressor response (Perry et al., 1970). Intra-arterial injection of low doses of cadmium (40 μg) produced a pressor response in rats. With higher dose (320 μg) intra-arterial injection of cadmium in rats produced initial depressor response followed by a transient pressor response (Perry and Yunice, 1965). Fadloun and Leach (1981a) showed similar depressor effects followed by pressor effects after intravenous administration of cadmium.

The acute pressor response in rats to intraperitoneal administration of cadmium was reported by Schroeder et al. (1966) and Perry and Erlanger (1971b, 1975). It has been shown that acute pressor response occurred within 1 minute after the intraperitoneal administration and persisted for several hours (Perry and Erlanger, 1971b, 1975). Hall and Nasseth (1980a), Hall and Hungerford (1982), confirmed that

1 mg/kg of cadmium intraperitoneally reliably elicited a pressor response in conscious rats under pentobarbital anaesthesia. ¼ of that amount gave maximal effect. Revis (1978) had shown a prompt increase in systolic pressure of about 38±5 mm Hg when cadmium chloride was given intraperitoneally to rats.

1.5.1.1. FACTORS MODIFYING THE HYPERTENSIVE EFFECTS OF CADMIUM :

1.5.1.1.1. Genetic factor :

Genetic variability has been discussed with multifactorial inheritance of vascular system sensitivity to cadmium (Ohanian et al., 1976, 1978, 1979). In experiments 1 to 16, week old, R & S rats of both sexes were injected two doses of cadmium (1 and 2 mg/kg body weight, i.p.), whereas controls received the same value of saline. Hypertensive renal vascular changes were observed in cadmium injected S (sensitive) rats but not in R (resistant) rats. The S females appear to be more sensitive than S males to the hypertensionogenic effects of cadmium. In another experiment groups of weanling female R & S rats were given 0, 1, 2.5, 5 or 10 mg cadmium/litre in drinking water and fed either low salt (0.4% NaCl) or high salt 4% NaCl for 28 weeks. Cadmium produced cardiac hypertrophy (1 mg Cd/litre) and hypertension associated with renal vascular

changes (1-5 mg Cd/litre) and it enhanced proteinuria (1-10 mg Cd/litre) in S rats on a low salt diet. Also the development of salt induced hypertension was accelerated in Cd fed (1 to 2.5 mg/rat) S rats. These adverse effects were not detected in R rats on either salt diet. Cadmium concentration in the kidney and liver of S rats were higher ($P < 0.001$) than the R rats. From these data it is indicated that the genetic difference influenced the pathogenesis of cadmium induced hypertension.

1.5.1.1.2. Influence of diet, sex and strain on cadmium-induced hypertension :

There was marked difference in blood pressure response to Cd with different forms of diet. Perry and Erlanger (1982) used three groups of rats which received low cadmium rye-based diet, composed of 60% rye meal, 30% powdered skim milk, 9% corn oil, 1% NaCl plus iron and vitamins; they also received the doubly deionized water fortified with 1 ppm Mo, 1 ppm Co, 5 ppm Cu, 10 ppm Mn and 50 ppm Zn. The other 6 groups were given a stock diet of Purina Rodent Lab Chow (Ralston Purina Co., St. Louis, MO). Three of the stock diet fed groups were given the same deionized and fortified water as the group receiving rye-based diet and three received doubly deionized water with no added metal. One of the three groups with similar food and water (rye

diet plus fortified water; stock diet plus fortified water, stock diet and non-fortified water) was not given cadmium in their drinking water, one received 0.1 ppm of Cd and one received 1 ppm of cadmium. It was found that the rats which were given rye-based diet and 0.1 to 1.0 ppm of cadmium regularly exhibited an average increase in systolic pressure of 15 to 20 mm Hg. For those rats given fortified water but stock diet instead of the rye-based diet, the increase in pressure induced by 0.1 or 1 ppm of cadmium were reduced or absent. More precisely for 0.1 ppm cadmium, the pressure increase was reduced by half after 3 months, by three-fourths after 6 months and it was entirely absent after 10 and 14 months. Doyle et al. (1975) found that 5 ppm cadmium in water did not induce hypertension in female Sprague-Dawley rats given a diet based on glucose and egg white. Eakin et al. (1980) also found that 10 and 20 ppm cadmium failed to induce hypertension in male OSU Brown rats given a glucose-casein diet that contained 0.1 ppm cadmium, 10 ppm of copper, and 0.2 ppm selenium. Although these workers observed no cadmium-induced increase in pressure, their control rats had a 30 mm Hg increase in systolic pressure, during 2-year observation period. Lener and Bibr (1970) reported that cadmium did not induce hypertension in female Wistar rats fed a commercial diet containing 0.04 ppm cadmium. Frickenhaus et al. (1976) reported that 20 or 40 ppm

cadmium added to food did not induce hypertension in Wistar rats fed commercial diet.

Petering et al. (1979) and Boscolo et al. (1981) found that 10 or 20 ppm cadmium for 9 months and 10 or 20 ppm Cd for 5 months respectively induced hypertension in male Sprague-Dawley rats fed Purina Rodent Lab Chow. Petering's stock diet-fed animals exposed to cadmium were affected in a way that was maximal in the beginning and decreased with time. Both Boscolo's and Petering's group used males whereas Perry and Erlanger (1982) used females. Further Perry et al. (1979, 1980) have shown that cadmium can induce comparable hypertension in both males and females, and the pressor effect is the same for the indirectly measured systolic and diastolic pressures. Ohanian and Iwai (1979) found that 1 ppm cadmium induced hypertension in hypertension-sensitive Dahl-rats fed the Agway diet, which contained 0.08 ppm cadmium and 16 ppm copper. Finally Walkar and Moses (1979) found an increasing pressor effect among rats given Purina Rodent Lab Chow and 10 ppm cadmium during 7-9 months of exposure. Schroeder and Vinton (1962) have shown difference in response to cadmium among the male and female. The mean systolic blood pressure of all of the female rats given cadmium (5 ppm) were higher than 170 mm Hg. Contrary to the females, male rats exhibited only a slight tendency to elevated systolic pressure.

Petering et al. (1979) could not produce hypertension in female rats and hypertension was observed only in male rats when cadmium was given orally for 36 weeks.

Various strain of rats have been used to induce hypertension by cadmium. Schroeder and Vinton (1962) used weanling rats (27-29 days old) of Long-Evans strain of both sexes. Perry et al. (1977) used female Long-Evans rats to induce hypertension with different dose of cadmium. All further work on hypertension in rats was carried out by Perry et al. (1971, 1977, 1979) in Long-Evans rats. Perry et al. (1979) tested the cadmium (5 ppm) in male Long-Evans rats and female Sprague-Dawley rats. After a year of exposure cadmium induced a typical increase of 17 mm Hg in males. The data shows that in Sprague-Dawley rats also, cadmium could induce hypertension. The Table VII shows the influence of sex, strain, diet and water on cadmium-induced hypertension on rats.

1.5.1.1.3. <u>The influence of cadmium on renal ischemic and salt-induced hypertensive animals</u> :

Schroeder et al. (1966) have shown that a synergistic hypertensive response was evident when small doses of cadmium were given intraperitoneally in renal ischemic rats. To ascertain whether or not cadmium is an accessory factor in

experimental renal hypertension, rats were subjected to partial constriction of the left renal artery (Schroeder et al;,1968b). From this study it was shown that in renal ischemic rats, the percentage of success and the levels attained were greater when cadmium (5 ppm) was also fed. It was further postulated that in the presence of renal ischemia, the ischemic kidney loses zinc and cannot readily sequester cadmium. In that event cadmium absorbed from water acts peripherally within a short time both enhancing the hypertension which had developed and initiating it when it had not.

Perry and Erlanger (1980, 1981) studied the average systolic pressure at various times for rats with a temporary excess of Na and/or continuous cadmium exposure. It was found that adding excess salt to Cd had little additional effect.

Hall and Nasseth (1979) studied the effect of cadmium on salt hypertensive rats. Young female rats were given a regimen of intraperitoneal cadmium treatments. They were sensitized to the development of salt hypertension by removal of one kidney and then given 1% saline solution to drink. It was further noted that over a five-week period the experimental animals consistently drank more saline than control. The systolic blood pressure of the cadmium treated animals remained normotensive in the salt hypertensive animals; whereas the control animals reached the hypertensive range (Hall and Nasseth, 1979).

1.5.1.1.4. Interaction with other metals :

The effects of 4 other metals such as lead, copper, selenium, and zinc on cadmium-induced hypertension were explored (Perry and Erlanger, 1977, 1978; Perry et al., 1974, 1980, 1983). At 3 and again at 9 months, 5 ppm Pb alone, i.e., without cadmium, increased the systolic blood pressure approximately as much as 5 ppm Cd. When Pb and Cd were given together, their individual effects on systolic pressure were found to be additive. Thus after 3 months Pb raised the average systolic pressure 14 mm Hg cadmium raised it 10 and Pb and Cd raised it 29 mm Hg. After 9 months corresponding values were 13, 17 and 26 mm Hg. Perry and Erlanger (1978) further extended their observation with lower doses of cadmium (0.1, 1 or 5 ppm) from the time of weanling and continued as long as 18 months when lead and cadmium together usually doubled the pressor effect observed with either alone. Exposure to 1 ppm of cadmium and lead for 3 months produced an average increase of 43 mm Hg in systolic pressure and therefore very significant hypertension for many animals. Perry et al. (1983) have recently shown that one per cent salt, 1 ppm cadmium or 1 ppm cadmium plus 1 ppm lead in drinking water caused similar mild hypertension in rats. The hypertensive effect of salt given for 4 months beginning at weanling, disappeared when the salt was

withdrawn but subsequently returned without further exposure. Rats continuously given 1 ppm cadmium during and after salt exposure were continuously hypertensive but salt did not increase their hypertension. Rats exposed to cadmium or cadmium plus lead for months remained hypertensive after the metal exposure was discontinued; addition of 0.35 ppm selenium corrected the hypertension in cadmium-fed rats, but had little effect in the cadmium plus lead exposed rats (Perry et al., 1983).

The situation with respect to copper (Cu) was different. After 3 months' exposure to copper (20 ppm) alone raised the average systolic pressure as much as 5 ppm Cd (Perry and Erlanger, 1977). However, at 9 months, Cu alone was no longer pressor but it completely blocked the pressor effect of Cd. At 9 month, none of the copper plus Cd had hypertension. Perry and Erlanger (1977) studied the effect of another metal, selenium, on cadmium-induced hypertension in rats. At 6 months both 0.9 and 3.6 ppm selenium alone tend to raise the systolic pressure; at the lower concentration the increase was marginally significant with $P < 0.05$. When combined with twice as much cadmium (2.5 and 10 ppm) both concentrations of selenium completely inhibited the cadmium induced increase in systolic pressure. In an earlier report Perry et al. (1974) attempted to inhibit the induction of hypertension by Cd. Two inhibitors were tested. Selenium, a

reported cadmium antagonist in several biological systems, and hard water, a general protective effect in human cardiovascular disease. In two sets of experiments with inhibitors, weanling Long-Evans rats were followed for a year in a low contamination environment and given a low Cd diet plus supplements of five essential metals with or without Cd in their only available drinking water. Two doses of cadmium were used either 2.5 or 10 ppm. There was significant increase in systolic pressure following cadmium alone. This increase in pressure disappeared when Se as the selenite was added in an amount equal to half the molar Cd concentration. Further, when cadmium was dissolved in hard water, hypertensive effect was not obscured. Both Se and hard water continued to inhibit the induction of hypertension (Perry et al., 1974). Perry and Erlanger (1977) studied the effect of zinc on the cadmium-induced hypertension. After 6 months of exposure 200 ppm Zn alone induced a marginally significant increase in systolic pressure, while 100 ppm Zn had no obvious effect on pressure. Both concentrations, however, apparently blocked pressor effect of both 2.5 and 10 ppm Cd.

When hydralazine was given in combination with Cd the hypertension induced by Cd was largely inhibited at 3 months while at 9 months there was a marginally significant increase in systolic pressure (Perry and Erlanger, 1977).

From this study it is concluded that cadmium induced hypertension was inhibited by selenium, zinc, copper and hard water. However, lead augmented the cadmium induced hypertension.

1.5.2. HUMAN DATA :

To put the available rat data into the context of human disease, the minimum pressor dose of chronically fed cadmium in the rat approximates 5 µg/kg/day (Perry et al., 1979). This is equivalent to 350 µg/day for a 70 kg man (Perry and Kopp, 1983). The average person in an industrial society is estimated to ingest 50 to 70 µg of Cd daily and certain high cadmium foods will raise this average markedly. Moreover, an average of 0.1 to 0.2 µg of cadmium is inhaled when cigarette is smoked with 25 to 50% of the inhaled cadmium apparently retained in the body (Menden et al., 1972). Although the kidney may not be the critical target organ with low dose cadmium exposure (Kopp et al., 1982) renal cadmium concentration in rats with cadmium induced hypertension bracket the renal cadmium concentrations in the average human being without specific recognized cadmium exposure (1 to 40 µg/g wet weight for hypertensive rats versus 14 to 28 µg/g for the average man) (Perry et al., 1977; Cherry, 1981). It is emphasized that human beings with toxic exposure to cadmium e.g. itai-itai disease or industrial poisoning are

not hypertensive and animals apparently showed the same phenomenon. Thus rats with toxic exposure have depressed rather than elevated cadmium level (Perry et al., 1977).

It has proved difficult to approach the problem of whether environmental cadmium exposure which is universal, although of variable intensity, plays any part in human essential hypertension which is presumed to have many causes and which occurs, albeit usually with only minimal elevation in blood pressure in about one American or European adult in three. The continuing accumulation of cadmium in human kidneys during childhood and early adult life produces a near maximal renal cadmium concentration during the decade of the thirties when most essential hypertension appears; however, there is little direct evidence relating the cadmium to the hypertension. In retrospect, early reports that very severe hypertensive subjects excreted an average of fifty times as much urinary excretion as normotensive controls (Perry and Schroeder, 1955) probably indicated rapid loss of renal parenchyma in combination with life time accumulation of cadmium which was sequestered there; likewise the return of the urinary cadmium concentrations towards but not to normal with partial control of the hypertension (Schroeder, 1965) is consistent with lessening the rate of renal destruction by antihypertensive treatment.

To date neither plasma nor urinary cadmium concentrations have been shown to be good indices of body burden. Ingested or inhaled cadmium present in plasma is presumed to be there only temporarily; its concentration may be related to recent exposure, but it seems unlikely to be a good measure of long time exposure. Indirect evidence has been interpreted as suggesting that urinary cadmium concentration is related to body burden of cadmium (Friberg, 1974) but there is no definitive evidence for this relationship, and high urinary cadmium levels may be more closely related to the loss of renal mass with its accumulated cadmium burden.

Usable information on body burden of cadmium has come only from assay of tissue obtained at autopsy, and there is considerable disagreement about the value of limited amount of available data collected in this manner. There are major obstacles to obtaining samples from appropriate hypertensive subjects and from a matched normotensive population (Perry and Kopp, 1983). On the one hand, subjects who have easily recognizable but advanced hypertension with diminished renal function are not appropriate since their original renal parenchyma, along with its accumulated cadmium has been largely replaced by scar tissue. On the other hand, in the ideal population with early hypertension, there are no reliable postmortem indices by which hypertension can be identified and blood pressure measurements are often

unavailable for subjects dying sudden and unexpected deaths (Perry and Kopp, 1983). Thus automobile accidents, which have been widely used as a source of subjects, may not provide a valid sample of the population, since they are weighted heavily towards those with a particular life style, e.g., young smokers who drive too fast, drink considerable alcohol, and may use other drugs as well. Finally since cigarettes are a major source of body cadmium (Lewis et al., 1972), it would simplify things to deal entirely with smokers or non-smokers, however, smoking history is frequently unavailable. With the foregoing reservations, most of the limited post mortem human data suggest that hypertensive subjects may have more cadmium in their kidneys than the normotensive matches. Thus five small series reported significantly more renal cadmium in hypertensives (Schroeder, 1965; Karlicek et al., 1971; Lener and Bibr, 1971; Perry and Kopp, 1983). One reported the reverse (Ostergaard, 1977) and Three found no significant difference (Morgan, 1952; Indraprasit et al., 1974; Syversen et al., 1976). Despite the small number of subjects per group and the wide individual variations, there is some agreement among the available series with respect to renal cadmium concentration. Thus the average renal cadmium concentrations of normotensive subjects varied from 21 to 29 µg/g wet weight in six of the nine series and the comparable averages for hypertensive

subjects ranged from 36 to 51 ug/g in five of the nine series (Cherry, 1981). One carefully studied series appeared to be a real exception with the hypertensive kidneys having significantly less renal cadmium than normotensive kidneys (average concentration of 12 versus 21 ug/g) (Ostergaard, 1977). When 70 hypertensive patients and 70 controls, matched for age and sex were investigated for blood cadmium, no significant difference between the two groups were detected although blood Cd level was significantly higher in smokers as compared to non-smokers (Beevers et al., 1976). In another study (Carruthers and Smith, 1979) on 22 of 31 residents of Somerset village where soil levels of cadmium were high had raised blood cadmium level and some had hypertension and renal tubular damage. Glausar et al. (1976) have found that untreated hypertensive humans had a significant increase in the blood cadmium level as compared with the normotensive humans. Hypertensive patients have been found to have increased urinary excretion of cadmium (Mackenzie and Kay, 1973). Dally et al. (1978) could not show any significant increase in blood cadmium level in hypertensive smokers as well as hypertensive non-smokers.

Debates on hypertension often result in a chicken-and-egg argument as workers try to dissect cause and effect with cadmium, if the animal results have relevance to man, there is evidence for a causal relationship with raised

blood pressure. However, several hurdles thus remain to be overcome before the element can be incriminated in hypertension. Even such evidence as is forthcoming cadmium can be only one additional factor in multifactorial condition. The days of hunting for a single cause of hypertension are (unfortunately) over.

1.6. IN VITRO PHARMACOLOGICAL STUDIES OF CADMIUM :

The effect of cadmium on various isolated tissues has been carried out by different workers. Perry et al. (1967) studied the effect of cadmium on isolated rabbit aorta and it was found that cadmium (1×10^{-4}M) reduced the contractile response to adrenaline (Adr). The response to angiotensin was increased after the incubation of the rat aorta with cadmium. On the other hand, it was found that reactivity of helically cut strips of aorta of Cd^{++}-hypertensive rabbit revealed decrease responsiveness to angiotensin (ANG) but not to noradrenaline (NA) (Thind et al., 1970a). It was further reported that Cd^{++} applied *in vitro* to aortic strip produced decreased responsiveness to NA, Adr, ANG and K^+ (Thind et al., 1970b). These authors postulated that this inhibitory effect could be associated with the binding of Cd^{++} by sulphydryl group of contractile protein. Toda (1973a) had shown that cadmium ions in concentrations of 0.02 and 0.1 mM decreased the response to K^+ in helically cut strips of aorta of rabbit.

The dose response curve of K^+ was significantly moved to the right following treatment with 0.02 mM Cd^{++} and markedly moved downwards and to the right by 0.1 mM Cd^{++}. Further reduction in the effect of NA was elicited by 0.1 mM Cd^{++}. However, the effect of NA was more resistant to Cd^{++} than that of K^+ (Toda, 1973a). The contractile responses to K^+ and Ba^{++} were inhibited by Cd^{++} in concentration higher than 0.02 mM whereas the responses to NA, histamine, and ANG were inhibited at higher than 0.1 mM. These results are different from those reported by Thind (1970a) who found that Cd^{++} was a more potent inhibitor of NA than K^+. Toda (1973a) suggested that cadmium interferes with the movement of Ca^{++} across the cell membrane. It was further shown that the inhibition by cadmium on the contractile response to stimulating agents was antagonised by cysteine. Further, Toda (1973b) had shown that in isolated left atria of rabbit Cd^{++} (0.02-0.5 mM) caused a marked depression of the action potential and a slight decrease in the resting potential. In more than half of the atria exposed to 0.5 mM Cd^{++}, action potentials were totally abolished with a slight reduction of the resting potential. The inhibitory effect of Cd^{++} on the isolated rabbit atria was partially reversed by cysteine, excess calcium, and ethylene glycol bis (aminoethyl ether) N,N,N,N-

tetraacetic acid. It has been demonstrated that Cd^{++} produces effects on the electrical and mechanical activity of intact isolated frog (Kleinfeld et al., 1955), dog (Kleinfeld et al., 1966) and rat heart (Kleinfeld and Stein, 1968). The major changes consist of a shortening of the action potential duration, a reduction in action potential amplitude, a decrease in contractility and a prolongation of P-Q interval in the electrocardiogram. It was reported that the induced change in electrical activity were not reversed by cysteine (0.005 - 0.01 ng/ml) (Kleinfeld, 1955). Kleinfeld and Stein (1968) showed that the cardiac contractility depressed by Cd^{++} was reversed by increasing the concentration of Ca^{++} in the bathing medium. In the rabbit sinoatrial (SA) node pace-maker fibres, the maximal diastolic potential and the threshold potential were decreased by Cd (0.1 and 0.5 mM) (Toda, 1973c). It was further suggested that the slope of diastolic depolarisation was decreased which results in bradycardia. In most preparations exposed to 0.5 mM Cd^{++} pace-maker activities were totally abolished (Toda, 1973c). The inhibitory effects of Cd^{++} were reversed by cysteine. The positive chronotropic effects of sympathetic nerve stimulation, NA, and histamine was inhibited by 0.02 and 0.1 mM of cadmium (Toda, 1973c).

In isolated frog sciatic nerve - gastrocnemius muscle

preparation exposure for 60 minutes to Cd^{++} (0.1 - 2 mM) caused a marked attenuation of the twitch tension developed by the stimulation of nerve but no or only a slight inhibition of tension produced by direct stimulation at maximum twitch height (Toda, 1976). It was further shown that the inhibitory effect was partially reversed by 1 mM cysteine. In frog sartorius muscle the addition of cadmium Cd^{2+} (0.1 mM) also abolished the end plate potential evoked by nerve stimulation but did not suppress potential changes induced by iontophoretically applied acetylcholine (Toda, 1976). The contractile responses of frog rectus abdominus muscle to acetylcholine were not significantly affected by cadmium (Toda, 1976). It was further shown that the impulse conduction along sciatic nerve was not impaired by Cd^{2+} but was by procaine. It was suggested (Toda, 1976) that Cd^{2+} interferes with the release of acetylcholine from the motor nerve terminals by reducing the transmembrane influx of Ca^{2+}. In isolated rat diaphragm Cd^{++} in concentrations of 0.1 and 0.5 mM caused a dose related inhibition of twitch tension developed by nerve stimulation (Usui and Toda, 1976). Twitches induced by direct stimulation were also significantly attenuated; however, attenuation was markedly less than that seen in the response to nerve stimulation.

As in the case of frog sciatic nerve - gastrocnemius preparation Cd^{++} appears to interfere with the release of

acetylcholine from the motor nerve terminals in rat phrenic diaphragm preparations by attenuating transmembrane influx of Ca^{++} (Usui and Toda, 1976).

In isolated seminal vesicle from guinea pig, the influence of Cd^{++} on the action of various smooth muscle contractile agents was investigated. The low concentration of Cd^{++} inhibited selectively the contraction by K^{+} and Ca^{++}, and high one also inhibited the contraction by acetylcholine, NA and barium in addition (Suzuki et al., 1977, 1978).

In helically cut strips of canine cerebral arteries exposed to Ca^{2+} free media depolarized by K^{+} the addition of Ca^{2+} caused a biphasic (transient and sustained) contraction while in coronary and mesenteric arteries Ca^{2+} produced a sustained contraction sometimes preceded by a slight transient contraction. These calcium induced contractions were attenuated by Cd^{2+} (5 to 100 μM) in a dose dependent manner, the attenuation being greater in cerebral than in coronary and mesenteric arteries (Hayashi and Toda, 1977).

Williams et al. (1978) have shown the enhancement of pressor response to nerve stimulation and NA administration in isolated perfused rabbit ear arteries. Cadmium in concentration of 0.075-0.25 μM causes enhancement of the pressor response to nerve stimulation but higher

dose (0.25 μM) caused inhibition of the response. To inhibit the pressor response to NA it requires 100 times higher concentration of cadmium than that needed for the inhibition of response to nerve stimulation. Animals treated chronically with low dose of cadmium (4-7 doses of 0.19-0.5 mg/kg evenly spaced over 1-2 weeks) were tested for the sensitivity to NA in rat aorta and rat tail arteries. Aorta from animals exposed to cadmium were more sensitive to NA than were aortas removed from control animals (Nechay et al., 1978). In addition, electrical stimulation of perfused tail arteries produced greater constriction in vessels removed from cadmium-exposed animals than in those from controls.

In vitro sensitivity to NA, KCl and electrical stimulation of portal and vas deferens was carried out in rats chronically treated with cadmium (5, 12.5, and 25 ppm) for one month (Fadloun and Leach, 1980). Cd pretreatment altered the response of portal vein to perimural stimulation. The nature of response depended on the frequency used; the response to 1 to 3 Hz were potentiated while response to 12 to 25 Hz were reduced. Cd^{++} (12.5 and 25 ppm) also enhanced the contraction to K^{+}, NA, and perimural stimulation in rat vas deferens. Niwa et al. (1981) studied the effect of different concentrations of cadmium in isolated guinea pig vas deferens with various agonists such as KCl,

NA, acetylcholine, barium and calcium. It was found that lower concentrations of cadmium (1×10^{-9}, 1×10^{-8} and 1×10^{-7} g/ml) enhanced the sensitivity of KCl, NA, barium, and calcium, whereas higher concentrations (1×10^{-6} and 1×10^{-5} g/ml) inhibited the responses to these agonists in this tissue. In the intestinal smooth muscle of guinea pig ileum, cadmium is shown to produce inhibitory effect to various agonists (Triggle et al., 1975). Asai et al. (1982) studied the effect of low concentration of $CdCl_2$ (1×10^{-8}M to 1×10^{-4}M) on the longitudinal muscle of the guinea pig ileum; and it was found that $CdCl_2$ caused a transient contraction. The dose-response curve was bell-shaped. Niwa and Suzuki (1982) recently studied the effect of low concentration of cadmium on rat aorta. It was found that cadmium produced contraction at low concentration but relaxation in high concentration. It was further established that low concentration of cadmium potentiated the effects of K^+, Ba^{++} and NA and high concentration inhibited the effects of these agonists (Niwa and Suzuki, 1982).

1.7. BIOCHEMICAL EFFECTS OF CADMIUM :

The observation of resulting hyperglycemia and glycosuria in cadmium-treated animals (Ghafghazi and Mennear, 1973) encouraged studies on carbohydrate metabolism in liver and kidney cortex. Cadmium exposure produced histopathologi-

cal changes, decreased glycogen levels and aldolase activity as well as an increase in hepatic phosphorylase - a enzyme (Stowe et al., 1972). Elevated levels of blood glucose and urea, and decreased contents of hepatic glycogen were observed in cadmium chloride-exposed adult as well as in neonate animals (Merali et al., 1975; Merali and Singhal, 1980). In the same set of experiments, the levels of hepatic gluconeogenic enzymes were increased in a dose-dependent manner. More pronounced changes were observed in various parameters in the neonatal rats exposed to cadmium from birth. However, withdrawal of cadmium exposure for 2 weeks could not restore the altered biochemical changes to normal limits (Singhal et al., 1974). Merali et al. (1975) found elevated levels of hepatic adenylate cyclase and cyclic AMP in cadmium-treated animals which may lead to stimulation of gluconeogenic enzymes. Rastogi and Singhal (1975) reported increased synthesis of adrenal catecholamines responsible for activation of enzyme phosphorylase, resulting in enhanced conversion of glycogen stores into glucose. Further, Ghafghazi and Mennear (1973) suggested the association of hyperglycemia with pancreatic function as they observed reduced glucose tolerance, serum immunoreactive insulin concentration and insulogenic indices in cadmium-treated mice. Merali and Singhal (1975) reported an increase in blood glucose and a decrease in resting insulin

levels as well as in glucose-stimulated serum immunoreactive insulin levels in cadmium-exposed rats. Furthermore, a smaller increase in phentolamine-induced immunoreactive insulin levels in metal-exposed animals compared with that of control animals indicated that reduced functional activity of pancreas was not the result of increased catecholamine release alone. A decreased *in vitro* release of insulin in response to high glucose concentrations was observed in islets isolated from cadmium-treated rats (Merali and Singhal, 1980).

In experimental animals, cadmium produces very profound effects on testicular tissue. It is believed that certain biochemical changes precede and may be responsible for the increased vascular permeability which leads to hemorrhagic necrosis in the testes. Lee and Dixon (1973) reported reduced male fertility and thymidine uptake into spermatogonial cells after lower level cadmium exposure of animals. Cadmium altered cyclic AMP metabolism in both primary and secondary sex organs, and the changes persisted even 28 days after the withdrawal of metal exposure (Sutherland et al., 1974). It is known that cyclic nucleotides play an important role in the process of reproduction through hormonal regulation. However, the contribution of cadmium-induced changes in cyclic AMP metabolism is difficult to assess at present. Although, experimental data show a great deal of

vulnerability of the male reproductive system to cadmium toxicity, the testicular necrosis (except for some unspecific histologic changes in the testes of some postmortem cases of industrial workers) has not been seen in humans so far.

Both zinc (Parizek, 1957) and selenium (Kar et al., 1960) seem to protect or reverse the cadmium-induced toxic manifestations in animals. Merali and Singhal (1975) reported that both selenium and zinc are effective in protecting animals against cadmium-induced alterations in glucose tolerance and insulin release responses to a glucose load. Further, pretreatment with zinc completely prevented the biochemical and functional damage of spermatogenic cells by cadmium. Some workers believe that the capability of zinc to induce metallothionein production protects the tissue from deleterious effects of cadmium by providing more binding sites. It is also postulated that since zinc is essential for various normal tissue functions, its displacement by cadmium may impair zinc-mediated metabolism leading to the injury of the tissue. Furthermore, the administration of higher doses of zinc may recapture its own sites by displacing cadmium, thereby protecting tissue from damage. The protective action of selenium may be due to the formation of a temporary complex with cadmium (Gunn et al., 1966) and leaving insufficient cadmium to act on other cellular constituents.

It has been found that neonatal animals are more susceptible to the neurotoxic effects of this metal compared with adults. Rohrer et al. (1978) reported changes in cerebral vasculature of the fetus exposed to cadmium. Cadmium accumulates in adult brain although it penetrates the blood-brain barrier with more ease in fetal rats. Greater concentrations of cadmium were found in brains of newborn rats after intravenous administration indicating that the blood-brain barrier to cadmium is not fully developed in the neonates (Wong and Klaassen, 1981). Rozear et al. (1971) reported decreased spontaneous neural firing after administration of the heavy metal into cerebral cortex or brain stem of cat. Further, in vitro addition of this metal was found to act as a synaptic blocking agent at both adrenergic and cholinergic synapses (Cooper et al., 1978).

Abnormal behaviour patterns were observed in fishes, bass and bluegill, exposed to cadmium through water (Cearley and Coleman, 1974). Pfister et al. (1978) treated adult rats with a single intraperitoneal injection of cadmium and found a significant decrease in open-field exploratory behaviour and rearing responses for up to 9 days following treatment. However, they did not find any behavioural disorder in the rats chronically treated for 3 months to 1 year. Smith et al. (1982) observed increased locomotor activity and better performance in learning and reversal learning trials in

cadmium-treated rats compared with controls. Wong and Klaassen (1982) showed histopathological damage and hyperactivity in newborn rats treated with this heavy metal. Attempts have been made to correlate behavioural changes with neurochemical alterations in specific brain regions. Rastogi et al. (1977) observed increased spontaneous locomotor activity coupled with increased levels of dopamine and norepinephrine in certain brain areas of rats chronically exposed to cadmium via drinking water before and after weaning. Ribas-Ozonas et al. (1974) reported an increase in 5-hydroxytryptamine and 5-hydroxyindole acetic acid in different regions of rat brain following 60 min of a single intraventricular injection of cadmium. Shukla and Singhal (1984) have shown that the administration of 0.5 mg Cd^{2+}/kg/day intraperitoneally to 22-day-old rats for 30 days was found to increase NA in the midbrain and pon-medulla, and 5-HT in the hypothalamus region without affecting dopamine levels. However, a dose of 0.1 mg Cd^{2+}/kg did not produce any change in the levels of monoamines but decreased GABA contents in the cerebellum and hypothalamus regions (Shukla and Singhal, 1984). In contrast, the intraperitoneal administration of 0.5 mg Cd^{2+}/kg/day to the adult rats has been reported to increase dopamine and decrease 5-HT levels in the whole brain (Shukla and Chandra, 1981). Further, *in vitro* addition of cadmium inhibited synaptosomal uptake

of ^{3}H dopamine and ^{3}H norepinephrine in a concentration-dependent manner (Hobson et al., 1983). Exposure to cadmium has also been found to alter the behavioural responses to apomorphine in rats suggesting the existence of dopamine supersensitivity in metal-exposed animals (Smith et al., 1983). It was proposed that the cadmium-induced increase in spontaneous locomotor activity may be related to increased adrenergic activity. However, the precise mechanism by which cadmium alters behaviour and produces neurotoxic manifestations remains to be elucidated.

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