SUMMARY

INTRODUCTION:

a) General:

The term 'pesticides' includes a large variety of compounds having diverse chemical nature and biological activity; group together, only on the basis of that use to destroy or eliminate pests. Pesticides have made a great impact on human health, production, and preservation of food, fibre, and many other cash crops. With the introduction of pesticides, farm practices have undergone revolutionary changes leading to incredible possibility. Under the U.S. Federal Environmental Pesticide Control Act the term 'pesticide' has been define to include any substance or mixture of substances intended for preventing, destroying, repelling or mitigating any pests like insect, rodent, nematode, weeds and other forms of terrestrial or aquatic plant or animal life or virus, bacteria and micro organism. Pesticide also includes substances or mixture of substances intended for use as a plant regulator, defoliator or desiccant.
Pesticides have given rise to many serious problems with a shading chill in the spine. Most of the chlorinated nondegradable pesticides leave residues in various living systems for prolonged periods of their life span, and are presumably responsible for a variety of known and unknown toxic symptoms. On long-term exposure, the pesticides are known to affect the immune system and cause cancer in both animals and human beings.

b) Pyrethrin:

Pyrethrin has been used for the control of harmful insect for many centuries. Its commonly applied to the dried flower heads of some species of Chrysanthemum like *C. cinerariaefolium*, *C. roseum*, *C. corneum* etc. Pyrethrin refers to the mixed active ingredients as present in commercially available extract of pyrethrum. Such extract contains about 30% by weight a mixture of 6 compounds in about the following amounts: pyrethrin I (11.4%), cinerin I (2.2%), jasmolin I (12%), pyrethrin II (10.5%), cinerin II (3.5%) and jasmolin II (1.2%). Since, the pyrethrins and cinerins are esters, they rather easily decompose in the presence of moisture and acids or alkalies.

Pyrethrin is used to control pest, in store food, against household and industrial pests. Pyrethrin aerosol sprays are excellent home insecticide because of their safety and rapid action, and it is also used in pest control of food and vegetables. Pyrethrin is commonly used for eradication of mosquitoes, in mats, coils etc.
c) Fernoxone:

Fernoxone is one of most widely used herbicide in India. It is also a growth regulator herbicide with auxin like activity. The common name of fernoxone is 2,4-dichlorophenoxyacetic acid (2,4-D). The chemical structure of 2,4-D resembles indole acetic acid, a naturally occurring hormone produced by plants to regulate their own growth. It is a broad leaf herbicide with systemic effect. All many forms or derivatives are esters, amine and salts of 2,4-D. These are formulated with solvents, carriers or surfactants, and are marked in the form of dusts, granules, emulsions, oils, water solutions, gels, and water-soluble packets in a wide range of concentrations. The herbicide, fernoxone or 2,4-dichlorophenoxyacetic acid (2,4-D) has been widely used for broad leaf weed control in farming, forestry, power line maintenance, aquatics, on home, and for other purposes.

For the present work, 2 weeks old sexually mature albino rats weighing about 120-150g. were injected intraperitonealy with different doses of pyrethrins and fernoxone, separately. LD_{100}, LD_{50}, and sublethal doses of these pesticides were determined. Sublethal doses of pyrethrin were 150mg/kg and fernoxone 220 mg/kg were used for experimentals. These above mentioned amount and doses of pyrethrin and fernoxone respectively were used as safe doses. Through out the experiments the rats were injected interperitonealy.
Through alternately in right and left sides of mid abdominal cavity on every 2nd day upto 1,5, 10, 20, 40, 80, 120 and 150 days to group of albino rats of almost same age. Each group was consisting of 6 healthy young rats and weighing approximately around same weights.

A group of 6 albino rats taken as control was injected with an equivalent volume of solvent only i.e. 10ml. petroleum ether and distilled water. Rat for control to pyrethrin and 5ml. distilled water/rat for fernoxone. In treated rats, 10 ml. of pyrethrin with petroleum ether and distilled water and 5 ml. of fernoxone with distilled water were injected. Various parameters of the rats treated with pyrethrin and fernoxone are studied. The Serum Glutamate Oxalate Transaminase (SGOT), Serum Glutamate Pyruvate Transaminase (SGPT), Alkaline Phosphatase, and Lactate Dehydrogenase (LDH) contents are analyzed by Computerized Biomedical Technology while cholesterol, and glucose contents are analyzed spectrophotometrically by Liberman's Burchards method in the blood on 1, 5, 10, 20, 40, 80, 120 and 150 days after treatment to albino rats.

Brain, thyroid gland, adrenal gland and pancreas (mixed gland) of the albino rats are described on the basis of histopathological analysis after microtomy and staining the sections with hematoxylin and eosin. The behavioral changes in albino rats are also studied, immediately after perfusing the drugs upto one hour continuously and than intermittently.
Behavioral changes:

Due to the effect of pyrethrin, there is an increase in locomotion like jumping, running etc. for about 20 to 30 minutes. But after a period of 40 days locomotory activity gradually slowed down and a delayed response to food and water was prominent. Neurotransmission is also affected because of pyrethrin. The prolong effects proved to be deleterious because of the accumulation of unmetabolised pyrethrin in the central nervous system (CNS), inhibition of certain function of the nervous system and formations of lesions in the nervous tissues.

With fernoxone, animals exhibited activity opposite to what it showed with pyrethrin, the experimental rat slowly ceased most of their spontaneous activities, and sit still, however they were observed awake and alert. Hind limbs of rat are usually more affected then the fore limbs. Some times circling, telepsy, loss of weight and negative geotaxis were observed in the treated animals. Fernoxone affects the normal neurology, furcuting, neuromotor reflexes and spontaneous motor activity.

Serum Glutamate Oxalate Transaminase (SGOT) and Serum Glutamate Pyruvate Transaminase (SGPT):

Increased SGOT and SGPT values clearly indicate that the pyrethrin has deleterious effect on liver and causes damage to hepatocytes. The
concentration of SGOT varies in control group in a definite range but recorded increased in treated rats. In the case of control group the concentration of SGOT remains between 114.75 U/L to 119.45 U/L upto 150th day, and in case of treated group upto the 5th day pyrethrin doesn't shows any significant increases in the level of SGOT but from 10th day level of the enzyme increases significantly.

The concentration of SGPT in control group ranges between 32.65 U/L to 36.25 U/L, and in the treated group though the level increases after the first day of treatment yet the significant changes appear only after 20th day of treatment. The highest average value recorded on 150th day is more than 3 fold of the normal value.

Fenoxone slightly increases but gradually. The value of SGOT from ranges from 149.65 U/L to 212.65 U/L, its value increases slowly and on 150th day of treatment it is recorded the highest (212.65 U/L).

The level of SGPT in the treated group decreases from 1st day to 40th day but from the 40th day onwards to 150th day its value increases from 69.05 U/L to 12.15 U/L. From these results it is concluded that level of SGOT increased high as compare to SGPT and the compounds have detrimental effects on liver.
Alkaline phosphatase:

Pyrethrin and like substances cause much more damage to liver, and are responsible for the elevated level of alkaline phosphatase. In healthy albino rat, its level ranges from 1.25 U/L to 2.96 U/L. By the exposure of pyrethrin the enzyme level increases from 5th day it is 4.74 U/L. On 150th day the enzyme level becomes just double than the amount in control healthy rats.

Fenoxone affects cell membrane permeability of hepatocytes that leads to leakage of alkaline phosphatase into the blood and hence increases its level. In experimental group there is a fluctuation (rise and fall) in the level of this enzyme, upto 10th day then its amount increases from 10th day to 20th day, from 120th day onwards it decreases and its value comes down to reach up to 3.05 U/L on 150th day. In treated animal its value increases from 3.38 U/L to 5.23 U/L (1st day to 150th day).

Lactate Dehydrogenase (LDH):

Minimum range of LDH in healthy untreated rat is between 50.6 U/L to 63.8 U/L. In treated rats, a significant increase of lactate dehydrogenase is observed from 10th day. The highest value of lactate dehydrogenase (LDH) is 106.25 U/L recorded on 150th day with the exposure of pyrethrin. The changes in the level of blood lactate on one hand may be associated with muscles
activity, myotonia, and impaired glucose metabolism leading to more synthesis of lactic acid in muscles thereby causing the more synthesis of LDH and to the other hand can be interpreted with hepatocytosolic release of LDH due to chemical caused injury.

Fenoxone shoots up level of LDH in the albino rats from 88.20 U/L to 110.6 U/L. whereas in control group its value was recorded between 66.2 U/L to 71.25 U/L.

**Cholesterol:**

Concentration of cholesterol in experimental group shows fluctuation as it increases up to 5th day than a decrease in the value of cholesterol up to 20th day, which again is followed by an increase from 20th day to 150th day. The mean value of cholesterol in normal untreated albino rats is measured 97.62 mg/dl while in treated animal it was recorded around 130.60 mg/dl to 96.0 mg/dl.

Fenoxone shows fluctuation in control as well in treated group. In control group, the mean value of cholesterol on 1st day is 155.3 mg/dl, and the value of cholesterol 150th day is 136.70. In the case of treated group there is slightly increases up to 5th day then the value decreases from 5th day to 80th day and again the cholesterol value increases. The value of cholesterol on 1st day is 191.05 mg/dl, and on 150th day its value is 188.55 mg/dl.
Glucose:

Glucose concentration in control group ranges between 55.20mg/dl to 56.80 mg/dl during the period of experiment. In pyrethrin treated rats glucose elevates from 1st day (75.25 mg/dl), and found to be high (113.20 mg/dl) up to the last day of the experimentation. The concentration was about 3 to 4 times more than the normal values in untreated rats. The increased level of glucose cannot be attributed to one factor only but should be considered as a synergistic effects of more than one phenomena like β cells in pancreas show progressive degeneration with dose, and duration that causes deficiency of insulin, which may be responsible for hyperglycemic condition. As it is evident from enzymatic analysis that liver is damaged hence there is disturbance in general physiological and biochemical processes like glycogenolysis, neoglucogenesis and lipogenesis. Besides, the hyper activity of zona fasciculata of adrenal gland.

Glucose level in fenoxone treated rats does not show any definite pattern of rise and fall. A significant rise is observed from 1st day and from 10th day the level gradually declines and comes to almost normal after chronic treatment of 150th day. Glut-receptor in liver, muscles and kidney become hyperactive to absorb more glucose or increased muscle activity or low activity of glucagon, or inhibition of glucose absorption from intestine or increased lipogenesis may be reasons for change of glucose level to normal range.
Brain:

In the brain cerebral hemisphere, and cerebellum are much affected by pyrethrin. It seems that mylien sheath and nervous connective tissue are depleted. Ventricles are highly intruded by cellular debris. In many regions particularly near the posterior colliculi the cell infiltration is comparatively high. Vacuolization is prominent in cells that are ultimately lead to the formation of fissures at places. Some elongated cellular structures are formed by linear arrangement of glial cells. In pia mater at few places, blank spaces are present.

Ventricular epithelium in lower part of the duct of Sylvius is multilayer thick, and is found detached at many places on ventrolateral side. Axonal cells do not seem to be affected, but the connective tissue and oligodendrocytes are affected with chronic treatment of pyrethrin. Vacuolization occurred around the Purkinje cells. Lysis of connective tissue in the peripheral region of cerebellum is of high degree as compared to the central part from where different nuclei tracti are originating or passing. In some tracti vacuolization, fissure formation, and picnosis in the oligodendrocytes are visible. The superficial layers are lighter than the inner layer. Dura it mater seems having interrupted blood supply. Neuroglia including neurocytes is distracted. There
is an irregular disposition of Purkinje cells. The process of cell lysis causes to affects thickness of cerebral cortex.

In fernoxone treated animals vacuolization is prominent in the cerebellum. Vacuolation in neuroglia is observed and the cytoplasm becomes hypergranulated. The Purkinje cells of cerebellum also show slight vacuolation. The normal disposition of neurons found to be disturbed by the action of fernoxone. Neuropathological changes like the development of lesion, atrophy of grey mater and possible disturbance in relay system of different part of the brain are observed in cerebral membrane. There is an apparent change in the behaviour. In the cerebellum negligible number of vacuoles are detected. Only at scanty places depression or thinning of neuroglia is observed. Cytoplasm is highly granulated with Nissl's bodies distributed at places. At few places, hypertrophy of nucleus is also visible. Neurocytes of medulla oblangata located in upper motor portion are found to be degenerated when exposed to 2,4-D for a long time. Accumulation of homovanillic acid, 5-hydroxy-3indole acetic acid and metabolites of dopamine and serotinin in brain are accumulated due to the disturbance in the metabolism of this neurotransmitter, when rats exposed to these poisons for a long period.
Thyroid:

With pyrethrin hypertrophy of follicular cells of thyroid is observed. Some squamous cells and less granulated cuboidal cells were noticeable at places in follicles. No lesion and hyperplasia is observed in the gland. Epithelium becomes vacuolated, and lobules are irregularly distributed. Parenchymal tissue of the gland surrounded by wide lymphatic capillary. The single layer of epithelium cells surrounding the central mass of gelatinous colloidal material, which is stained is moderately granulated. Follicular cells have many lysosomes and multi vesicular bodies.

In fernoxone treated rats the variation in the size of the cell is not much than that studied in the control normal rat. There is lesser damage to the follicular cells of thyroid. The very high concentration of fernoxone is found to cause hypertrophy of thyroid with depletion of colloid. Parafollicular cells are scattered in connective tissue. Epithelium is observed with vascularized basement membrane.

Adrenal:

In rats treated with pyrethrin for long period, cells of adrenal gland develop vacuolization in the cytoplasm. Hypertrophy of the cortical region is observed and ultimately lesions are visible at some places. No vacuolization in
zona reticularis and glomerulosa is seen but disturbed pattern of cell was also marked. Hyperplasia in adrenaly medullary cells was observed.

By the chronic treatment of fernoxone large vaculation in the cytoplasm, picnosis of nuclei degeneration of mitochondria, deposition of fat and atrophy of cells are observed. Here also, disturbed pattern of cells is noticed. Hypertrophy was observed in the cells of zona glomerulosa of the adrenal cortex. At some places only little cytoplasm in few cells are observed.

**Pancreas:**

Pyrethrin causes no pathological changes either in exocrine, aciner cells or endocrine, islets of Langerhens. Normal distribution of zyonogen granules in pyramidal cells was observed. In β cells little but uniform distribution of granules and was seen. Inter aciner alveolar tissue is normal with usual blood vessels and fiber distribution.

In fernoxone treated rat, small vacuolization develop in the cytoplasm of aciner cells. β cells and other cells of islet of Langerhans were reduced in number. In β cell the granules are comparatively low in number and located near the periphery of cells, below the plasma membrane. At some places, hypertrophy and atrophy of the lobules was observed. Picnotic nuclei and hyperplasia of cells were observed. In β cells granules are scanty but no vacuolation could be detected.