CHAPTER FOUR

DISCUSSION

A. GENERAL DISCUSSION :-

Out of 15 toxicity tests considered today, the highest rated test include acute mortality. Acute tests were considered ecologicaly significant, most scientifically and legally, detensible, modest in predective capability, most simple and cost effective and were considered to have the greatest utility (Buikema et al.,1982).

The present investigation aims at the understanding of the effect of malathion and sumithion on fish gill and mortality by using acute toxicity test. The present investigation deals with LC 50 Values for C.punctatus, the histomorphological structure of a gill in control fish, histopathological changes in gill structure due to toxic effect of two pesticides and histochemistry of mucosubstances in control fish and fishes under experimental tests.

1. PHYSICO-CHEMICAL PROPERTIS AND LC 50 VALUES :-

Malathion and sumithion altered the physico-chemical properties of water in test aquaria containing different concentrations of both these pesticides. The change in the physico-chemical properties was due to different chemicals present in these pesticides. These chemicals alter the pH and DO of normal water and lead to water pollution. Hardness of water, however, was not altered at any concentration of

both the pesticides.

Eills (1937) and Verma and Dalela (1976) have noticed that pollutants affect the fish life through three different ways. i) By causing respiratory and circulatory failure through intereference with the respiratory functions of the gills, ii) By specific toxic action and absorption through gills or lining of mouth or other external structures, iii) By toxic action after absorption through the gastrointestinal tract.

In the present investigation the fish mortality of C.punctatus the different concentrations of malathion and sumithion was observed because of fluctuations in the values of parameters such as pH and DO. The main changes in the quality of water were, decrease in pH from 7.35 to 7.05 due to malathion and from 7.35 to 7.10 due to sumithion and decrease in DO from 8.5 ppm to 4.2 ppm for malathion and from 8.5 ppm to 5.5 ppm for sumithion. However, the hardness of water was remain unchanged. The pH and DO values from the experimental sets of aquaria at different concentrations of the malathion and sumithion showed direct correlation to each other and the values decreased with increase in the concentration of malathion Figs. 1st and 2nd and sumithion Figs. 3rd and 4th. These changes causing deterioration of water media in which the fishes live.

It is a common practice to use the LC 50 observed for short time period of exposure to determine

the safe concentration of a toxicant. Although this is of little practical significance, where exposure is likely to be prolonged. For any given period of exposure a harmful dose, as far as physiological, biological and behavioural aspects are concerned is much lower than the LC 50 (Fujiya, 1961) . The observations in the present investigation further support this.

The LC 50 values for number of pollutants have been estimated for variety of fishes by several workers earlier. The 96 hrs. LC 50 value was estimated, for three herbicides between 490-1220 mg/l by Woodward and Mayer Jr. (1978); in cutthroat trout, S.clarki for O_3 and Cl was 0.03 ppm and 0.07 ppm respectively in channel catfish, I.Punctatus by Hammond and Bishop (1979); for endosulfan was 35% in L.rohita by Rao et al. (1980); for selected rice pesticides to cray fish was 0.28 ppm by Chean et al. (1980); and 3.5 ± 0.2 ppb in M.aculeatus by Rao et al. (1981) and same for different acids was between 3.5 and 3.0 in bluegill sunfish, L.macrochirus by Ellagaard and Gilmore (1984). Whereas, 48 hrs LC 50 value for malathion was found 5.5 ppm in T.mossambica by Sailatha et al. (1981) and for metasytox (an insecticide) was 5 ppm in C.striatus by Natarajan (1982). Similarly LC 50 value for Cd of 216 hrs. was estimated to be 15 ppm by Chung (1983).

2. Fish behaviour :-

After the fishes acclimatized in normal tap

water, they were transfered to the aquaria containing different concentrations of malthion and sumithion. The changes in the behaviour showed by C.puntatus, used presently were more or less similar to that showed by other fishes exposed to other pollutants. The fishes showed sluggish and inactive movements and became diagonal in position and finally floated on the water surface. Due to continued treatment the fishes die because of suffocation and exert a synergistic effect after entering the gastrointestinal tract and fish shows asphyxia. Similar changes in the behaviour of T.mossambica and R.daniconius exposed to distillary effluent have been observed by Nikam (1986). Sukumar and PR. Karpagaganpathy (1988) observed erratic swimming, convolutions and hypersensitivity and finally death in freshwater C.lalia exposed to carbofuran. At sublethal exposure he observed hypersensitivity and jerkey movement, and finally acclimatization to pesticide environment. Similarly, Rajamanickam and PR. Karpagaganapathy (1988) noticed erratic swimming, convloution, spirraling, tremors, jerky movements, rapid operculum movements and surfacing activity in T.mossambica exposed to sublethal concentration of lindane.

B. HISTOLOGICAL DISCUSSION :-

Droscher (1882), Keis and Wilmer (1932), Bevelander (1935) and Carmignani and Zaccone (1974) in

comparitive study on the branchial epithelial cells of fishes from different habitats found several types of intraepithelial cells which belonged to unicellular, multicellular and transistional types. However, in present investigation the intraepithelial cells are found in the form of mucous cells and acidophil cells.

Droscher (1882) , Keis and Wilmer (1932), Bevelander (1935) and Munshi (1960, 64) reported on the occurance of bi-or trinucleated acidophil cells. In the present study uninucleated bi and trinucleated acidophil cells are found in the epithelium at interlamellar regions. Bird and Eble (1979) found chloride secreting cells clustered at the base of secondary lamellae in close proximity to afferent lamellar arteries. The present investigation also agreed with this view. Similarly the mast cells were reported amongst the gill epithelial cells Munshi (1964).

According to Munshi the mast cells which are the reserviors of heparin and histamine found in gill of fishes (1964, 1968 a, 1968 b, 1980). However, the presence of mast cells have been denied by Ingale (1981), in number of fishes he has studied. The present investigation is agreed with this view of Ingale.

Morgan and tovell (1973), Laurent and Dunel (1980), Lewis and Potter (1982) reported constant structure of branchial epithelium.

Hughes and Grimstone (1965), Munshi and Singh (1968 b), Hughes and Morgan (1973) and Hughes and Munshi (1979) reported on many structural changes in general, in the organization of the gill in relation to the enviornment. They considered that thickened epithelium could cut down diffusion of gas from the gills into the water and if the water is deoxygenated this might serve as an oxygen conserving device, by preventing the diffusion of oxygen from the blood into the water. The present investigation revealed three to four layered thick epithelium. The epithelium was more thick towards the tip of the primary gill lamellae and at their bases.

The results obtained with histological staining revealed presence of two rows of secondary gill lamellae on primary gill lamellae as transverse folds, which are thin filamentous structure present at the right angle to the axis of primary gill lamellae.

Each secondary gill lamellae consisted of a central core of blood sinuses lined and supported by pillar cells and covered by basement membrane and an outer epithelium. Similar histological studies have also been reported on this structure of the secondary gill lamellae which is common to all the fishes. Acrivo (1938); Hughes and Grimstone (1965); Munshi and Singh (1968 b); Hughes and Wright (1970); Hughes and Munshi (1973 a, 1973 b); Das and Srivastava (1978) and Munshi (1960, 1964, 1980), Ingale (1981) and Nikam (1986) in their studies described the pillar

cells. According to them, pillar cells are formed of a central cell body with wide flanger at each end spread out beneath the covering layer to meet the flanger of adjacent cell. The present investigation also agreed with this view.

The pillar cells were found to arrange one after the other in secondary gill lamellae in present study. However, there is controversy regarding the arrangement of pillar cells within the gill lamellae. Rauther (1937) found the arrnagement of pillar cells at random, Acrivo (1938) indicated the arrangement of pillar cells to be in parralel rows of ganoid fishes. Munshi (1960, 1961, 1962 a, 1962 b, 1968) and Munshi and Singh (1968 b) produce clear evidence about the arrangement of the pillar cells, in air breathing fishes. Furthermore, Munshi and Singh (1968 b) and Hughes and Munshi (1979) reported the absence of pillar cells in A.cuchia and M.cuchia. According to them in these fishes the blood capillaries are lined by endothelial cells. In most of other fishes the blood channels are formed by pillar cells except the marginal ones, which are lined by endothelial cells. Hughes and Munshi (1979) and Munshi (1980) stated that the columns of the basement membrane material runs transversely across the secondary lamellae connecting the basement membrane of two sides which lie deeply enfolded by pillar cells. They also added that these cells together with basement membrane form the structural

support of the secondary lamellae. This view has been supported by number of authers (Hughes and Grimstone, 1965; Hughes and Wright, 1970; Hughes and Weibel, 1972; Das and Srivastav, 1978). Mallatt and Paulsen (1986) stated that the pillar cells in hagfish gill lamellae contain bundles of microfilaments associated with collagen columns as in other fishes. However, their extracellular collagen columns are rare than in other fish gills.

Another important component of fish gill are the mucous cells. Droscher (1882), Keis and Wilmer (1932), Bevelander (1935), Morgan and Tovell (1973), Carmiganani and Zaccone (1974) , Laurent and Dunel (1980), Lewis and Potter (1982), Ingale (1981) and Nikam (1986) reported the presence of considerable number of mucous cells together with respiratory epithellal cells in the fish gill. The present investigation also revealed presence of mucous cells along with the respiratory epithelial cells which show regular distribution along the portion of epithelium, corrosponding to the surface of branchial arch along the base of the interlamellar spaces. These are localised more in number towards the tip of primary gill lamellae.

Droscher (1882), Keis and Wilmer (1932), Bevelander (1935) and Carmignani and Zaccone (1974) stated that the mucous cells are some times rounded and some times goblet in the form with a nucleus that is not always identifiable because it is covered with a great

quantity of mucous material. This material appears in the form of lumps and shows no affinity for eosin. Mucous cells were also reported in the secondary lamellae of gill, in number of fishes by Zoccone (1972); Sing and Thakur (1975), Gona (1979 a, 1979 b) and Ingale (1981). This may be related to more secretion of the mucus in diverse ecological conditions to which the fishes are subjected.

Droscher (1882), Owen (1886) and Keis and Wilmer (1932) identified the presence of chloride cells in the gill epithelium of fishes. According to them the chloride cells are nothing but mucous cells since they actively secrete chloride ions. Saxena (1966), Czolowska (1966), Guha et al. (1967), Hamada (1967), Singh (1968 a), Zaugg (1970), Zaccone (1972), Carmignani and Zaccone (1974), Munshi (1974), Singh and Thakur (1975), Das and Srivastav (1978) and Ingale (1981) stated that the number of chloride cells is the specificity of the habitat. All are of the view that the number of chloride cells is more in fresh water fishes. Although the fishes from diverse habitat have not been investigated presently it is clear that the gills of the fish which has studied presently shows acidophils (chloride cells,) localised in the epithelium in between the primary gill lamellae and the epithelium lining the primary gill lamellae. Keis and Wilmer (1932), Virbhadrachari (1961), Vicker's (1961), Das and Srivastava (1978), and Munshi (1980) are of the

agreement that according to changes in the salinity some of the mucous cells are transformed into chloride cells for the transport of electrolytes. Virbhadrachari (1961) observed change in the number of mucous cells in Etropus with variation in the salinity. Further , he stated that some of the mucous cells get transformed into chloride cells and this may be the reason why there is a change in the number of mucous cells. This type of study is also carried out by Fletcher et al. (1976), Ojha and Munshi (1974) and found similar results. The present investigation reveals only few number of acidophils (chloride cells) as compared to the mucous cells.

C. HISTOPATHOLOGICAL DISCUSSION :-

The gills of fishes come in direct contact with chemical and physical irritants in the surrounding water. (e.g. various toxicants, extremes of temperature or pH) These pollutants cause several types of gill lesions. In fish, the pharynx is a major site through which waterborne pollutants can enter the body, and gills are often among the organs most affected by such compounds. (Lemke and Mount, 1963; Lang, 1967; Skidmore and Tovell, 1972; Smith and Piper, 1972; Abel, 1974; Wobeser, 1975 a; Rombough and Garside, 1977).

The literature has generated a point of controversy. Some studies have detected no gill lesions resulting from exposure to irritant (Crandall and

Goodnight, 1963; Herbert and Shurben, 1964; Lowe, 1967; Walsh and Ribelin, 1975; Schreck and Lorz, 1978 ; Daoust and Ferguson, 1984). These studies involved the full range of exposure conditions (acute, chronic lethal, chronic sublethal) and toxicants classes (heavy metals, ammonia, organic materials). These authers have claimed that the widely reported gill alterations are primarily artifacts. However, most of other studies revealed that the most of the reported lesions are real and these are only some alternate views.

Furthermore, many authors have noted that under any given set of exposure conditions, each kind of gill lesion tends to vary widely in intensity. The different gills and lamellae within a single fish tend to show differing severity of alterations (focal lesions; Kuhn and Koecke, 1956; Christie and Battle, 1968; Gardner and Yevich, 1970; Temmink et al., 1983). Different fish also tend to be affected in varing degrees (Van Valin et al.,1968).

Before about 1970, it was hopped that irritant-induced changes in fish gill structure would prove to be irritant specific and pathognomonic (Sprague, 1971;). The authors of recent partial surveys of the literature, however, have concluded that similar branchial changes are induced by a range of different irritant substances (Pauley and Nakatani, 1967; Benville et al., 1968; Flis, 1968 b, Skidmore and Tovell, 1972; Matthiessen and Brafield, 1973; Dimichele and Taylor, 1978; Jauch, 1978; Mitchell et al., 1978; Dalela et al ., 1979; Katz,

1979; Wedemeyer et al., 1979; Scott and Rogers, 1980; Haensly et al., 1982; Solangi and Overstreet, 1982).

The most commonly recorded alteration is a lifting of epithelial cells from the gill lamellae and the interlamellar zones of the gill filament representing an infilteration of this epithelium with fluid.

The lifting of epithelial cells from the underlying tissue has been reported in rainbow trout exposed to anionic detergent (Abel and Skidmore, 1975), in gills of I.metas due to Cu treatment (Baxter and Cirrie, 1978), in C.guchua exposed to acute and sub-acute levels of endosulfan and rogar (Dalela et al, 1980), in juvenile and different developing stages of S.gairdneri exposed to rogar, benomyl, pomuran and domatol (Sigloch, 1981), exposed to petroleum (Engelhardt et al., 1981), exposed to sublethal concentration of Zn for 96 hours (Tuurala and Soivio , 1981), exposed to sublethal concentration of permethrin, an insecticide (Kumaraguru et al., 1982); in freshwater T.fasciatus exposed to sublethal and lethal concentration of BHC (Gupta, 1982) and in N.notopterus exposed to sublethal concentration of dinitrophenol and pentacholorophenol individually and their combinations (Gupta, 1986). Lifting of epithelial cells have also been reported by , Yang and Sen (1986) due to acute toxicity of ammonia on common carp, C.Carpio, by Solangi and

Overstreet (1982), due to acute toxicity of ammonia on common carp, C.carpio and Englhardt et al., (1981) on S.gairdneri due to petroleum exposure.

Skidmore and Tovell (1972) and Rombough and Garside (1977) also reported the lifting of epithelial cells from the gill lamellae and interlamellar zones of the gill filament. The results obtained in the present investigation indicated the lifting of inter-lamellar epithelium from basement lamina in the gills of fishes exposed to 10 ppm malation and 10 ppm, 20 ppm and 25 ppm sumithion. The lifting of epithelial cells from the secondary gill lamellae was noticed in the gills of fishes exposed to 12 ppm. malathion and 25 ppm sumithion.

The other common lesions include the hypertrophy, hyperplasia and rupture of branchial epithelium, lamellar fusion, bulging of lamellae (clavate-globate lamellae: Eller, 1975), hypresecretion and proliferation of mucous cells and changes in chloride cells and gill vasculature.

Hypertrophy of the secondary gill lamellar epithelial cells have been reported in S.gairdneri due to the effects of dehydroabietic acid (Tuurala and Sovio, 1981). The hypertrophied epithelial cells and mucous cells have been reported in the trout exposed to ammonia for about 12 months (Smith and Piper, 1972; Cruz et al., 1982); in milk fish C.chanos due to acute exposure of ammonia (Cruz et al.,1982), in esturine fishes exposed to crude

oil for 30 days (Solangi and Overstreet, 1982); in Juvenile channel cat fish, I.punctatus due to sewage effluent (Mitz and Giesy, 1985) and in common carp, C.carpio exposed to acute toxicity of ammonia (Yang and Sen 1986).

The histopathological results obtained in the present study indicated the hypertrophied mucous cells in the gills of fishes exposed to 6 ppm, 8 ppm malathion and 10 ppm and 15 ppm sumithion.

The hyperplasia of the epithelial cells lining the secondary gill lamellae and of interlamellar epithelium is also most commonly reported gill lesions.

Hyperplasia of the epithelial cells in the gills of trout exposed to ammonia for about 12 weeks have been reported by Smith and Piper (1972) and Cruz et al., 1982. Similar observations have been reported by number of investigators. Sigloch (1981) has been reported on the sever hyperplasia in juvenile S.gairdneri, C.carpio, I.indus and different stages of S.gairdneri due to rogar, benomyl, pomuran and domatol. Kumaraguru et al. (1982) reported on mucous cell hyperplasia, hyperplasia and fusion of adjacent secondary lamellae in gills of S.gairdneri exposed to permethrin (Pesticide). Hyperplasia have also been reported in milk fish, C.chanos exposed to acute concentration of ammonia, by Cruz et al., (1982) , in Atlantic cod, C.morhua exposed to crude oil for about 12-13 weeks by Khan and Kiceniak (1984) , in juvenile cat fish, I.punctatus due to sewage effluent by Mitz and Giesy (1985), in B.stigma due to chronic exposure of fish to endosulfan, malathion and sevin

by Khillare and Wagh (1988).

The present investigation also shows the hyperplasia of the epithelial cells of secondary gill lamellae in gills of fishes exposed to 4 ppm, 6 ppm, malathion and 10 ppm sumithion. The present investigation also revealed thickening of the interlamellar epithelium in the gills of fishes exposed to 4 ppm, 6 ppm and 8 ppm malathion and 10 ppm, 15 ppm, 20 ppm and 25 ppm sumithion.

The histopathological results obtained in the present investigation on gills of C.punctatus indicated the fusion of the secondary gill lamellae in the fishes exposed to 4 ppm, 6 ppm, 8 ppm malathion and only 10 ppm sumithion. Similar histopathological lesions have been reported by number of investigators. Smith and Piper (1972) reported fusion of gill lamellae in the gills of trout exposed to ammonia. Similar histopathological changes have been reported in the gills of C.gachua due to toxic effect of rogar and endosulfan (Daleb et al., 1980), rainbow trout, S.gairdneri from fresh water and marine water, exposed to petroleum (Engelhardt et al., 1981) and exposed to sublethal concentration of Zn for 96 hours (Tuurala and Sovio, 1981) in C.puctatus due to toxic effect of Zn (Khangarot, 1984), in rainbow trout, S.gairdneri exposed to permethrin (Kumaraguru et al., 1982) milk fish C.chanos due to acute exposure to ammonia (Cruz et al; 1982) trout exposed to ammonia for about 12 months (Cruz et al.,1982) , esturine fishes exposed to crude oil for about 30 days (Solangi and Overstreet, 1982), B.stigma exposed to dimethoate (Singh and PR. Karpagaganapathy, 1988).

Clavate-globate lamellae or lamellar aneurism have been reported less frequently. Blood filled aneurism have been reported in trout exposed to ammonia (Smith and Piper, 1972), in milk fish, C.chanos exposed to ammonia for about 12 months (Cruz et al., 1982). Similar histopathological alterations have also been obtained in the present study in fishes exposed to 10 ppm and 20 ppm sumithion only.

Necrosis in the gill tissues have been reported in C.gachua after acute and subacute exposure to endosulfan and rogar for 32 days (Dalela et al., 1980), in juvenile S.gairdneri, C.carpio, I.indus and developing stages of S.gairdneri due to acute toxic effect of rogar, benomyl, pomuran and domatol (Sigloch, 1981), in rainbow trout S.gairdneri exposed to permethrin (Kumarguru et al.,1982), in cat fish, M.vittatus exposed to sublethal and chronic levels of methylene blue for about 56 days (Ahmed, 1984), in C.striatus due to sublethal dose of sevin and thiodon (Jaikar and Kulshrestha, 1985), in N.notopterus exposed to sub-lethal concentration of phenol, dinitrophenol and pentachlorophenol individually and to their combination (Gupta, 1986), in common carp, C.carpio due to acute toxicity of ammonia (Yang and Sen, 1986) and in B.stigma due to chronic exposure to endosulfan, malathion and sevin for 16 weeks (Khillare and wagh, 1988). The present studies revealed the necrosis of the gill tissue in fishes exposed to 4 ppm, 10 ppm and 12 ppm

malathion and 30 ppm sumithion.

Histopathological results obtained in the present investigation revealed rupture of branchial epithelium in fishes exposed to 4 ppm, 6 ppm, 8 ppm, 10 ppm malathion and 25 ppm and 30 ppm sumithion. Similar histopathological changes have been reported in T.mossambica and R.daniconius exposed to distillary effluent (Nikam, 1986). Similarly rupture in the lamellar epithelium have also been reported in the S.trutta exposed to low dissolved O_2 (Drewett and Abel, 1983). They further reported the disarry of the secondary lamellae in the same fish exposed to lindane. Degeneration of the gill epithelium have been reported in brown trout due to lindane (Abel, 1976), in esturine fish, F.heteroclitus exposed to acute and chronic exposure of mercury (Koepp and Dozato, 1979), in fresh water teleost fish, O..punctatus exposed to sublethal concentration of diazinon (Sastry and Sharma, 1981), due to toxic effect of Zn (Khangaroot, 1982), in brown trout exposed to lindane (Drewett and Abel, 1983), and in C.carpio exposed to acutly toxic concentration of CH_3Br. (Segers et al., 1984)

The histopathological results in the present investigation revealed degeneration of the gill epithelial cells in fishes exposed to 4 ppm, 6 ppm, and 8 ppm malathion and 25 ppm and 30 ppm sumithion.

The some of the histopathological changes in the present investigation revealed dilation of blood spaces in

the secondary gill lamellae and/or primary gill lamellae (fishes exposed to all concentrations of malathion and sumithion). Similar histopathological results have also been reported in I.metas due to Cu treatment (Baxter and Cirrie, 1978), in S.gairdneri exposed to sublethal concentration of Zn for 96 hours (Tuurala and Sovio, 1981), in Atlantic cod C.morhua exposed to crude oil for 12 to 13 weeks (Khan and Kiceniuk, 1984) and in T.mossambica and R.daniconius (Nikam 1986) . These authors moreover reported the leucocytes accumulation in these dialated blood spaces. On the other hand Mallatt (1985) in his statistical review reported constriction of blood channels in the gills of some fishes induced by toxicants and other irritants. However, in the present investigation such a type of gill lesion has not been observed at any concentration of malathion and sumithion.

Mallatt (1985) in his statistical review stated that the curling and twisting or gill lamellae have been reported by Flis (1968 a, 1968 b), Skidmore and Tovell (1972), Kumar and Pant (1981). Such a lesions in the gill lamellae have also been reported by Nikam (1986) in two teleost fishes, T.mossambica and R.daniconius. The present investigation revealed the curling of secondary gill lamellae in the gills of fishes exposed to 6 ppm malathion and 10 ppm, 15 ppm, 20 ppm, 25 ppm sumithion.

Damage to pillar cells and marginal cells have been reported by Gupta (1986) in N.notopterus, following

exposure to phenolic compound. Damage of pillar cells have also been reported by Segers et al. (1984) in C.carpio exposed to acute toxicity of CH_3Br, by Nikam (1986) in T.mossambica and R.daniconius exposed to distillary effluent and by Singh and PR. Karpagaganapathy (1988) in B.stigma exposed to dimethoate. Schmid and Mann, 1961; Brown et al., 1968; Skidmore and Tovell, 1972; Abel & Skidmore, 1975; Abel, 1976; O.Conner et al., 1977; Rombough and Garside, 1977; and Dalela et al., 1979 also reported the similar histopathological alternations. According to Mallatt (1985) such vascular damage, however, usually was found only in animals exposed to very high does or to fishes that were near death. The present study revealed the damage of pillar cells in fishes exposed to 10 ppm and 12 ppm malathion and 25 ppm and 30 ppm sumithion.

Clumping of cells of cartilage axis have been reported by Gupta (1986) in N.notopterus following exposure to phenolic compounds. On the other hand thickening of gill rays have been reported by Singh and PR. Karpagaganapathy (1988) in B.stigma exposed to dimethoate. The present investigation also revealed thickening of gill rays in the fishes exposed to 4 ppm, 10 ppm malathion. On the other hand degeneration of central supporting material has been found in the present investigation. This observation is supported by the results, obtained by Nikam (1986) in T.mossambica and R.daniconius exposed to distillary effluent.

The number of mucous cells increases when fishes exposed to some toxicant atleast to sublethal exposure. Khan and Kiceniuk (1984) reported increased number of mucous producing epithelial cells in Atlantic cod, G.morhua exposed to water soluble fractions of Venezuelan and Hibernia crude oil. Similarly, Nikam (1986) also reported increased number of mucus cells in gills of T.mossambica and R.daniconius exposed to sublethal dose of spent wash. However, on the other hand loss of mucous goblet cells has been shown to occur in certain fishes due to higher concentrations of sea bloom (Shimada, 1983). Akitaka et al., 1982 reported the appearance of mucous cells besides chloride cells in the gills of yellow tail, S.quingueradita exposed to sea bloom. Nikam (1986) also reported decreased number of mucous cells in gills of T.mossambica and R.daniconius because of acute toxic effect of the spent wash. The present investigation also revealed increased number of mucous cells in the gills of present fish exposed to sublethal concentration of malathion and sumithion. However, the number of mucous cells decreases when the fishes exposed to lethal concentration of both the pesticides.

Koepp and Dozato (1979) reported epithelial damage and damage of chloride cells in the gills of F.heteroclitus due to acute and chronic exposure to mercury. According to them the epithelial cells damage was immediate for all concentrations, however, chloride cells damage occur later. The occurance of chloride cells has been established amply (Hughes, 1979; Munshi, 1980).

Engelhardt et al. (1981) recorded chloride cells abnormalities in the gills of S.gairdneri acclimated to fresh

water and marine water due to petroleum exposure.

Akitaka et al. (1982) observed chloride cells with many cellular extensions along with mucous cells in yellow tail, S.quingueradiata exposed to sea bloom. Krous et al. (1982) found 9.5 fold increase in the number of lamellar chloride cells in S.gairdneri when he was studing movement of nitrate across the gill epithelia.

Nikam (1986) also reported the damage or reduction in number of chloride cells in both the fishes he has studied. Similar observations using different fishes and employing other effluents have been obtained by Leino and McCormic (198

In the present investigation the chloride cells ar refered as acidophil cells. These cells found distributed i epithelium between the two primary filament and also in th interlamellar epithelium. The number of these cells found dec reased with increase in concentration of both the pesticides.

When the fish is exposed to any types of toxi material in the water, to avoid it, the fish gills and ski secrete the mucus material which gets spread over on genera body surface and thus, tries to have protection.(Matelev et a 1971; Lock and Overbreeake, 1981).

The present investigation revealed increase in th mucus secretion when the fishes are transfered to tes aquaria containing different concentrations of both th p ticides. The initial increase in the mucus secretion b both epithelial cells and mucous cells is important i this regard. As the experimental fishes need protecti from the toxic substances (malathion and sumithion naturally , the demand for additional mucous cells eith

have to increase in size or in number or both. As stated previously, it has been observed that the mucous cell number and/or size gets increased, in the gill epithelial cells of present fish, atleast in sublethal doses of both the pesticides.

The additional discharge of mucus forms a thick covering on gill epithelia and it interferes with its basic function of respiration. No doubt mucus in very small thickness (few microns only) is helpful in the process of respiration (Ingale, 1981 ; Haniffa and Sundaravadhavan, 1984). But as indicated earlier due to toxic effect of pesticides the thickness of it increases and hence, the hinderance in respiration.

The acidic medium has a corrosive action on the gill lamellae and this increases along with the increase in acidity. The decrease in pH means increase in the acidic medium. In the present work, it has been noted that the pH of water gets decreased with addition of both the pesticides (from 7.25 to 7.05 for malathion and from 7.32 to 7.10 for sumithion). To prevent from the harmful action of acidic medium the mucous cells secret additional mucus, which has been observed in the present work.

It has been postulated by Metelev et al. (1971) that n acid media the gill epithelia absorb the acidic solution. By doing so the blood picture of the fish gets ltered. This leads to the impairment of many blood functions. The combined effects, as indicated above,

induce coagulation of fibrin, haemolysis, clumping of blood cells, etc. In concept with these the pH of the blood gets altered (Metelev et al., 1971). When such impure blood goes to brain its function get severly affected. These effects are more pronounced in the respiratory and cardiac centres which ultimately lead to the slowing up of the above functions. It has been observed in the present investigation that, after the initial excitation, the fish became sluggish. Further, the sluggishness increases along with the concentration of both the pesticides and time. This must have been the result of the reduction in respiratory and cardiac activities. In extreme cases both these activities stop completely. Such total stoppage of respiration and cardiac functions have been amply illustrated by a number of investigators in different fishes exposed to various types of polluting agents (Metelev et al., 1971; Van Der putte, 1981).

The various histopatholegical lesions accured in the gill of C. punctatus due to malathion and sumithion intoxication and the possible reasoning for them will be discussed hereafter.

Lifting of epithelial cells from the gill lamellae and the interlamellar zones, of the gill filament which has been observed in the present investigation may be due to infiltration of this epithelium with fluid as stated previously by Skidmore and Tovell, 1972; Rombough and Garside, 1971).

Past authers have divided the commonly reported gill lesions into two groups hypothesized to indicate two different kinds of reaction to irritant. The first group of lesions consists of necrosis and rupture of the branchial epithelium. These alterations are belived to reflect "the direct deleterious effects " of irritants (Temminik et al ., 1983).

The necrosis and rupture of the branchial epithelium observed in the present investigation are also considered due to the direct deleterious effects of malathion and sumithion.

The present investigation, also revealed that the necrosis and rupture of branchial epithelium may be dose dependent as these are observed only under lethal concentrations of both the pesticides rather than under sublethal concentrations. Kuhn and Koecke,(1956)and Abel, (1976) reported that under the most highly toxic conditions, necrosis and rupture of the branchial epithelium are the only gill lesions that occur. Branchial cell death and rupture could develop via two different mechanisms, autolysis induced by the cell's own enzymes following the toxicant induced disruption of cell processes, or rapid lysis, caused by the direct lytic action of toxicant on cell constituents (Abel, 1976). In the present investigation, however, it may consider due to second type of mechanisms (i.e. due to direct lytic action of toxicant).

The second group of irritant-induced gill lesions include epithelial cell lifting, lamellar fusion, hypertrophy and hyperplasia of the epithelial cells, mucous hypersecretion, clavate-globate lamellae, congested blood cells in lamellae, mucous cell proliferation , leucocyte infiltration of gill epithelium, dilation or constriction of lamellar blood sinuses. According to Morgan and Tovell (1973) these lesions are produced as defense response of the fish rather than due to direct toxicant action. For example, mucous hypersecretion helps bar toxicant entry and chloride cell proliferation seems to reflect some role for chloride cells in toxicant extrusion or neutralization (Motais and Romeu, 1972; Matthiessen and Brafield, 1973; Calamari et al., 1980; Karnaky, 1980; Crespo et al., 1981; Oronsaye and Brafield, 1984; Mallatt et al., 1985).

Lifting, swelling and hyperplasia of the lamellar epithelium could serve a defense function, as these alterations increase the distance across which water borne irritants must diffuse to reach the blood stream, Lamellar fusion could be protective in that, it diminishes the amount of vulnerable gill surface area. Branchial response that serve to slow entry of toxicant have undesirable side effects of threatening to suffocate the fish (Skidmore, 1964; Burton et al., 1971). This may explain why the epithelium at the lamellar tips, where most respiratory exchange occurs, seems resistant to

irritant-induced change (Skidmore and Tovell, 1972; Morgan and Tovell, 1973; Abel and Skidmore, 1975; Roberts, 1974; Hughes et al., 1979; Segers et al., 1984).

The occurance of chloride cells (here referred to as the acidophils) has been estabilished amply (Hughes, 1979 and Munshi, 1980). They are known to be involved in the osmoregulatory functions. Because of the toxic action of both the pesticides, they either get damaged or reduced in number. Hence, the functions of these cells get impaired. Similar observations using different fishes and using other pollutants have been obtained by Leino and McCormic (1984), Nikam (1986). They are further of the view that the alterations in chloride cells are due to the change in the pH of medium. The present work also indicated the change in pH values towards acidic side after the addition of both the pesticides.

The present work revealed curling of the secondary lamellae at some concentration of malathion and sumithion. This may be due to the damage of pillar cells which normally support the gill epithelia and the vascular tissue in the secondary lamellae (Munshi, 1980; Haniffa and Sundaravadhanam, 1984). From the point of curling phenomenon of the secondary lamellae, the work of Skidmore and Tovell (1972) is important. They have shown that the pillar cells contract tremendously when exposed to polluting agents. Because of this, the hydrostatic

pressure in them is reduced that leads ultimately to curling up of the secondary lamellae, as observed in the present investigation.

D. HISTOCHEMICAL DISCUSSION :-

The present investigation was undertaken to augment our knowledge of characterisation of mucousubstances in branchial epithelial cells and mucous cells of C.punctatus before and after the exposure of different concentrations of malathion and sumithion and to compare the results obtained with that of existing literature.

Mucus is a product that results from an interaction between mucin, a glycoprotein containing secretion and water. In teleost fishes mucin is secreated by mucous cells which are found throughout the epidermis, the linings of the buccal cavity, the pharynx and the gills, as well as in epithelia of a variety of internal organs.

The histochemical results obtained in the present investigation revealed the absence of glycogen and any atypical mucosubstances in branchial epithelial cells or mucous cells in control fish. The absence of glycogen has also been reported in branchial epithelium of young individuals of T.marmorata and T.ocellata. Carmignani and Zaccone (1974). Parveen and Vasantha (1988) reported decrease in glycogen content in the gills of fishes

exposed to endosulfan. According to them the carbohydrates which are the readymade source of energy may be utilized under pesticide stress. Diwan et al. (1979) also reported the decreased level of glycogen in the gills of two fishes H.fossilis and C.bactrachus. However, in the present investigation the histochemical results indicated the absence of glycogen in epithelial cells and mucous cells of fishes exposed to different concentrations of malathion and sumithion.

Zaccone (1973) reported that during development of respiratory tract in M.sphenops a gradual increase occurs in the number of mucous elements in the epithelium of the oropharynx, branchial arches and branchial chamber with only small number in epithelium of the branchial filaments. During the development the quantity of mucopolysaccharides with carboxyl and phosphate groups in the mucus gradually increases and mucopolysaccharides with sulfated groups are always present in small quantities after birth. Porcelli and Novelli (1970) also reported the presence of sulfated mucosubstances in the muciparous cells of developing branchial epithelium of S.fario. In the present study, however, the histochemical characterisation of mucosubstances in the epithelial cells or mucous cells in the gills of present fish have not been studied during the developmental stages.

Cupurão (1967) reported the presence of sulfated mucopolysaccharides in the branchial epithelium

of X.maculatus. Cockson (1971) investigated the gills of T.Shirana Chilwae histochemically and found carboxylated mucopolysaccharides, however, sulfated mucopolysaccharides were found to be absent. They further reported the probable presence of the mucopolysaccharides, mucoprotein and protein carbohydrate complexes in the same fish.

Zaccone (1972) reported the presence of sulfated mucosubstances in the mucocytes of the branchial epithelium of M.cephalus and A.jordani (Selachian).

Carmiganani and Zaccone (1974) found considerable quantity of sulfated mucosubstances in the epithelial cells of the gills of adult specimens of T.mormorata and T.ocellata. They further stated that the neutral mucopolysaccharides are present only in the ytoplasm of the epithelial cells of the young forms, they are almost entirely absent in that of adult specimens. Similarly, the mucous cells show considerable quantity of sulphated mucopolysaccharides (Chondroiotin sulfate B). They reported the absence of hyaluronic acid and chondroiotin Sulfate A and C but presence of very small quantities of sialic acid. This facts indicated that the mucopolysaccharides elablorated in the young specimens, both by the epithelial cells and by the mucous cells and in adult specimens only by the mucous cells, is very similar. According to them this fact might also be related with the function of these substances with regards to the respiratory apparatus since the mucus is a typical

lubrificating substance; a considerble quantity of it would be produced by both types of cells for the greater protection of the branchial apparatus, espacially in the early periods of fuctioning of gills. Later, this lubrificating function is fulfilled only by the mucous cells.

Yamada and Yokote (1975) carried out orphological analysis of mucosubstances of some epithelial tissues of eel, A.Japonica. They reported that the mucosubstances of the gill are found in goblet cells which elaborate a neuraminic acid containing mucosaccharides with vicinal hydroxyl sulfate and carboxyl groupings and glycoprotein respectively. Ingale (1981) reported that the epithelical cells in fresh water fish like Kharpa, sheengata, katarna and shingti elaborate only neutral mucopolysaccharides. However, these cells in the remaining fresh water fishes (Katarna, Kolshi, murungi etc.) contains neutral mucopolysaccharides and sialic acid fraction. In esturine and marine fishes the epithelial cells elaborate additional mucopolysaccharides i.e. sulfated polyanions. He showed that the amount of the sulfated polyanions elaborated by marine fishes is more than those of esturine forms .

Furthermore Ingale (1981) described six types of mucous cells on the basis of nature of mucopolysaccharide elaborated by particular cell. He also pointed out that these mucous cells also showed distinct variations with

regard to their mucopolysaccharide elaboration. According to him M1-mucous cells are present in kharpa, katarna, vambat, murung parag, popat, mutri and marine catfish which elaborate only neutral mucopolysaccharides, M2-mucous cells are present in kolshi, popat, marine cat fish and pomphret elaborating only sulfated polyanions, M3-mucous cells secrete only sialic acid which are found in fishes like vambat, murungi, and parag, M4-type of mucous cells mostly found in esturine and marine water fishes and in few fresh water fishes which elaborate a mixture of neutral mucopolysaccharides and esters, M5-mucous cells are present in kolshi katama, shingti, sheengala, valshaveda, parag and marine cat fish. They elaborate a mixture of neutral and acidic muco polysaccharides (sulfate esters and sialic acid). Such cells are observed in all the fishes. He further added that irrespective of mucopolysaccharides elaboration by the mucous cells they show peculiar distribution in the various regions of the gill. From the above observations he concluded that the mucopolyscaccharide content of the various tissue components of the fish gill show distinct difference and this can be correlated with the type of habitat of the fish.

The present histochemical studies demonstrated the presence of neutral mucosubstances in the gill epithelial cells of control fish while only neutral mucosubstances in M1-type of mucous cells and only

sulfomucins in M2-type of mucous cells.

There are some reports on isolated mucopolysaccharides from the gill of fishes. Ahuja (1970) studied comparitive response of chloride cells and mucous cells to chloride and sulphate enriched media in the gill of Gumbusia and Catala. They stated that the presence of large number of chloride secretory cells and their significant hypertrophy are correlated with high tolerance of Gumbusia. They observed loss of mucous cells in the gills of Gumbusia adapted to active salts beyond 2.7% salinity.

Wasserman et al. (1972) isolated an acidic glyconsaminoglycans from the soft tissue of the gills of carps, C.carpio . According to them the major part of glyconsaminoglycans found in the gills of this fish is the mixture of chondroitin sulfates of various levels of sulfation and of glucosamine containing substances belonging to the heparin sulfate or heparin group. There is possibility that these polysaccharides may be the constituents of the connective tissue, mast cells, etc. They found anticoagulant activity in most of these fractions.

Some of the studies are concerned with the effects of some toxicants or change of habitat on mucin secretion in the gills of fishes.

Vinnikov et al. (1979) studied the mucus from the respiratory tract (gill) of A.guoldenstaedti and

A.stellatus under electron microscope and found the presence of secretory granules, 0.4-1.0 μ in diameter. According to them the mucus contain significant amounts of Na^+, K^+ and Mg^{++}. They made comparison between the concentration of these 3 electrolytes in the mucus from the gills of fresh water fishes and fishes from sea water and found no significant difference between electrolyte content of gill mucus.

However, in the present investigation isolation and seperation of mucosubstances from the gill have not been done and only histochemical localization of mucosubstances have been carried out.

Narsimm and Parvatheswarao (1974) reported that the neutral mucopolysaccharides were the most abundant type in the gill of T.mossambica. They found increased quantity of these mucopolysaccharides in the gill when these fishes acclimated upto 50% sea water and decresed quantity on further acclimation. By observing this they concluded that 50% sea water is iso-osmotic to the fish and the mucous cells in the gills were mostly in an immodified state. They further stated that the mucous cells in gills of these fishes acclimated to hyposmotic and hyperosmotic media may be modified in to chloride ecreting and excretory cells. Thus, the mucopolysaccharides may be concerned with ion fluxes and their quantity changes in the heterosmatic media may be governed by the combined effects of changes in ambient

salinity and in the blood medium osmotic gradient.

Bird and Eble (1979) stated that when juvenile and adult A.rostrata were adapted to freshwater, their gills exhibited staining properties similar to these of fresh water teleosts. According to them, the gill filaments of A.rostrata contained a centrally located cartilagenous gill ray which stained intensely in periodic acid Schiff (PAS) , Alcian blue at PH 0.5 and 5.7 with high concentrations of $MgCl_2$. They further stated that the chloride cells which were clustered at the bases of secondary lamellae contained high concentrations of carbohydrate polycarboxylates that stained intensely with PAS and Alcian blue at PH 2.6 . The mucous cells on the gill filament stained for acid mucopolysaccharides, and the epithelial basement membrane stained heavily for sulfated mucopolysaccharides.

Plonka and Neff (1969) analysed histochemically the nature of mucosubstances in the gills of 12 brook trout, S.fontinalis exposed to acidic water and made comparison with that of equal number of control trouts. They found an increase in mucin content and proliferation of mucous cells between gill lamellae in gills of fishes exposed to acidic water.

Pism et al. (1980) examined the mucopolysaccharides in the chloride cells of M.capito and L.reticulatus adapted to fresh or salt water.

Chloride cells were characterised by a high concentration of polysaccharides in their apical region (at the vesiculotubular system) and by a special polysaccharide cell coat. They stated that the polysaccharide originated from the golgi area by 12 hours and accumulated within the vesiculotubular system and were released in the apical cavity of the cell within 24 hours. The amount and turnover of polysaccharides were increased in fish adapted to salt water than in fresh water.

Effect of heavy metals on mucus secretion in teleost fishes have been reported by several investigators. Mucus accumulation on the gills of fishes has been observed in gold fish following exposure to lead nitrate (Westfal, 1945) or mercuric chloride (Muckone et al., 1971; Lock , 1975; Varanasi et al., 1975) in cat fish treted with copper-or zinc sulphate (Lewis and Lewis, 1971) and in rainbow trout exposed to methyl mercuric chloride (Olson et al., 1973; Lock, 1975)

Similarly, Lock and Van Overbeek (1981) studied the effects of mercuric chloride and methyl mercuric chloride on the activity of mucous cells in the gill epithelium of rainbow trout, S. gairdneri. They used 3 parameters to measure this activity. i.e. density of mucous cells, mucus content of the cells and the mucus release into the water. They found release of mucus into water is most sensitive to treatment with mercury. When the two compounds are used in equal concentrations of

mercury, mercuric chloride exerts a much stronger effect on mucus secretion than methyl mercuric chloride.

The results obtained in the present investigation also revealed increased amount of mucin secretion by epithelial cells and mucous cells in gills of fishes exposed to different concentrations of malathion and sumithion. The present study also revealed the change in the nature of mucosubstances secreted by epithelial cells in gill of fishes exposed to all concentrations of both the pesticides.

Some other studies have been carried out on the gill of fishes e.g. enzyme activity, which also deserve some attention in variety of fishes from diverse habitats and under experimental conditions, the brief review of which is taken in chapter introduction (Review of literature).

Singh (1972) studied the various chemical moieties by histochemical methods from the chloride cells of marine teleosts. The histochemical studies of these cells show that they were sudanophilic, indicating the occurance of phospolipids. Changes in lipid composition due to dodecylbenzenesulfonate in gill of round crucian carp has been studied by Nakanish (1986). Phleger (1975) have demonstrated the synthesis of lipid by A.rostrata gill.

There are few reports on protein content and their metabolites in gill of fishes. Bhaskar et al. (1982)

found elavated level of structural and soluble proteins in the gills of T.mossambica exposed to alkaline water while studing the protein metabolism in gill. Decreased level of NH_3 and urea has been reported in the gill of T.mossambica exposed to methyl parathion (Rao et al. , 1981) . According to them the change in NH_3 level in tissue suggest the possiblity of NH_3 recycling to counteract the methyl parathion toxic stress. Ghosh (1985) studied the effect of chromium on free amino acid content of gill in C.punctatus. They found increment in free amino acid content which may be due to enhanced proteolysis or/and due to stepped up protein synthesis. Gupta et al. (1987) investigated the effect of starvation on total protein and free amino acid in gill of C.punctatus and found decrease in total protein content while increase in amino acid content in gill.

Munshi (1980) is of view that the acidophil granular cells of the gill epithelia are diastase resistant, PAS positive while those belonging to the connective tissue system are PAS negative. However, he further claims that the granular cells become PAS positive after extraction of lipids. According to Munshi the acidophil cells do not show metachromasia with toludine blue but react strongly towards PAS reaction. The PAS positive material has been identified as mucoprotein complex as per the suggestion of Pearse (1961, 1968).

It is worth mentioning here that the mucosubstances, secreted by the gill epithelial cells and mucous cells perform some functions in the life of fishes. The fuctions performed by mucus secreted by epithelial cells; lining the gill, oropharynx, intestine epidermal cells of skin etc., have been stated by number of workers. According to Jakowaska (1963) continuous production and release of mucus could prevent the settling of pathogenic organisms on the body of fish. Fletcher and Grant (1969) stated that the presence of bacteriolytic enzymes, antibodies and lysosome activity in surface mucus indicate its protective function.

Histochemical studies by Asakawa (1970) and Bremer (1972) and biochemical studies by Enomoto and Tomiyasu (1962), Enomoto et al. (1966) and Lemoine and Olivereau (1971) on mucosubstances in the epidermis of the eel have shown that the properties of the mucosubstances are complex and diversified. Similarly, Bremer (1972) also stated that the histochemical properties of goblet cell mucosubstances are not necessarily uniform in the epidermis of fishes.

An investigation by Rosen and Conford (1971) demonstrated that mucus provides assistance in the fish's locomotion by strongly reducing the friction with water. Yamazaki (1972) stated that a layer of mucus prevents or reduces the skin damage caused by abrasia. A related function is the capacity of mucus to coagulate and

precipitate particles in suspension, thus providing protection to delicate tissue such as the gill filament. In addition, mucus is used by a number of teleosts for specialised functions related to their reproduction, e.g. nestbuilding and feeding of the youngs (Lock and VAN Overbeeke, 1981).

One of the more important function of the mucus is, its role in osmoregulation. It has long been discussed whether mucus affects the osmoregulatory capacity of teleosts (Van Oosten, 1957; Hughes and Wright, 1970). However, Maetz and Bornancin (1975) stated that the gills of teleosts play an essential role in osmoregulation . Similarly, Pickford et al., 1966; Potts and Evans, 1966; Wittouck, 1975; Marshall, 1976, Fletcher et al., 1976; Hentschel and Miller, 1979 also suggested the role played by mucus in osmoregulation. Cockson (1971) atributed the osmoregulatory role for carboxymucins in the gill epithelium of T.Shirana Chilwae. It has been suggested that the layer of mucus covering the organs like skin or gills may facilitate ion uptake by its ion-binding capacity (Kirschner, 1977; Marshall, 1978).

Narsimham and Parvateswarao (1975) reported that the mucous cells in gill of T.massambica acclimated to hyposmotic and hyperosmotic media may be modified into chloride secreting and excretory cells respectively, thus mucopolysaccharides in gill of T.massambica may be concerned with ion fluxes. Lock and VAN Overbeeke (1981) stated that the idea that the mucus affects the

osmoregulatory capacity of teleosts is not surprising as mucous cells abound at the sites where ionic interactions between the fish and its environment takes place; the gills, the skin the intestinal tract and, in some cases the kidney.

The present investigation is in aggreement with that which has been suggested by the earlier workers in this conncection.

The increased production of the mucus from gill epithelial cells and/or mucus cells, as well as hypertrophy of mucous cells and increased number of them in fishes exposed to some toxicants suggested the protective role played by mucin. Similarly, increased number of mucus cells in fresh water adapted fish than in seawater adapted fish and modification of mucous cells into chloride and excretory cells, with change in media may suggest the osmoregulatory role of the mucus in the fish life.

In the present investigation, increased production of mucus from both, epithelial cells and mucous cells in the present fish was found, with increase in the concentration of both the pesticides. This may be for the protection of gill epithelium from the damaging effect of malathion and sumithion. In addition, the increased level of mucin and hypertrophied and increased number of mucous cells for increased level of mucus secretion may indicate the protective role of mucosubstances. In the present

⊙ except 25 ppm sumithion, where sialomucins are also screated along with neutral and sulfomucins.

investigation the increased number of mucous cells for increased level of mucus secretion may indicate the protective role of mucosubstances. In the present investigation the increased number of mucous cells found at least at sublethal exposure of fishes for both the pesticides.

The present investigation revealed change in the nature of mucus secretion from only neutral in the epithelial cells of control fish to neutral and sulfomucins in gill epithelial cells of fish exposed to 4 ppm malathion, neutral and sialomucins in fish exposed to 6 ppm malathion; neutral, sulfo and sialomucins in fish exposed to 8 ppm malathion and neutral and sulfomucins in fishes exposed to remaining concentrations of malathion and all concentrations of sumithion.⊙ However, there was no change in the nature of mucosubstances secreated by the mucous cells even after the exposure of fishes to different concentrations of both the pesticides.

The problem why there is change in the nature of epithelial secretion, is not clearly understood. However, it is assumed that by doing this the fishes may tried to protect themselves from the dangerous effect of the pesticides.

In similar studies, increased amount of mucin secretion have been reported by some earlier workers. Plonka and Neff (1969) found an increase in muc[illegible] conte[illegible]

and proliferation of mucous cells in twelve brook trout, S.fontinalis exposed to acidic water. Lock and VAN Overbeeke (1981) found increased release of mucus in water by the gill epithelium of rainbow trout exposed to methyl chloride, and methyl mercuric chloride.