Chapter 10

Ions and Associated ATPases
INTRODUCTION:

The osmotic and ionic characteristics of the body fluids and tissues of freshwater organisms are largely influenced by the ambient medium. The maintenance of the homeostasis in such a condition is very much dependent on the osmoregulatory properties and it is a vital phenomenon to maintain the physiological balance between the external environment and internal milieu of an animal. Alteration in osmotic regulatory mechanism under toxic conditions may cause severe imbalance in biochemical composition of the tissue fluids followed by undesirable metabolic consequences. The principal components of osmoregulation are ions. The ions help in maintenance of perfect osmolarity of the cell.

The inorganic ions play an important role in osmotic phenomena and in the regulation of cellular metabolism. These are required by all animals to provide suitable medium for protoplasmic activity. Any imbalance in the levels of these ions in animals will lead to impairment in various physiological activities (Leone and Ochs, 1978; Baskin et al., 1981). Freshwater fishes are hyperosmotic to their medium. They gain water osmotically and tend to loose solutes by diffusion. In the regulation of osmolarity of a system, sodium, potassium and calcium ions play a significant role to keep the hyperosmotic properties of these animals (Narasimhan et al., 1983).

Sodium (Na⁺) is the principal cation of extracellular fluids of most animals. It maintains electroneutrality and internal sodium concentrations (Maetz and Romu, 1964). It also plays an important role in osmotic regulation of body fluids.
and also serves as an essential activating ion for specific enzyme systems. The increased sodium content may cause a shift in ionic symmetry with a consequent change in membrane permeability and functional efficiency of Na$^+$ - K$^+$ pumps.

Potassium (K$^+$) is the prominent intracellular cation of animals and is an important cofactor in the osmotic pressure regulation and acid-base balance (Saxena, 1957). Potassium activates certain enzymes (transferases) and is critical for the maintenance of normal membrane excitability. The consistancy of intracellular potassium, even with varying total osmotic concentration of habitat, may represent a very old cellular character (Prosser, 1973). It plays an important role as an osmotic inorganic effector in animals (Lagerspetz and Senius, 1979).

Calcium (Ca$^{2+}$) is another important osmotic effector and is involved in conferring stability to the cell membrane. It is also a cofactor for several oxidoreductases, proteases, ATPases and couples the oxidation with contraction in muscles. For the maintenance of structural integrity of mitochondria, sarcoplasmic reticulum and rate of enzyme catalysis, calcium content of the tissues is an important factor (Harper, 1985). Calcium is a general regulator of permeability of cell membranes to water and other ions. High calcium level generally decreases permeability and low calcium increases it. Hence, calcium level can be taken as an index of mitochondrial integrity and cellular metabolism (Chan et al., 1980). Any change in calcium level can alter the mitochondrial function, protein synthesis and steady state of enzymatic reactions (Narasimha Reddy et al., 1979).

All these ions exist in bound as well as in free forms. Bound ionic forms involve
in metabolic functions, and free ions involve in osmolarity in order to contribute to homeostasis of the cell system.

Adenosine triphosphatase (ATPase) enzymes are vital for regulating oxidative phosphorylation, ionic transport, muscle function and several other membrane transport dependent phenomena. Na\(^+\) - K\(^+\) adenosine triphosphatase (ATPase) has a central role in branchial transepithelial ion transportation in fishes (Epstien et al., 1980). This enzyme is present in the cell membrane of virtually all vertebrates (Post and Sen, 1967; Skou, 1975) and is particularly abundant in tissues associated with ionic and osmotic regulation (Boonkoom and Alvarado, 1971). Mg\(^2+\) ATPase is a mitochondrial enzyme involved not only in the lysis of ATP but also have a significant role in the initiation of ATP synthesis (Lehninger, 1979). Mg\(^2+\) ATPase is found in association with both Na\(^+\) - K\(^+\) and Na\(^+\) - NH\(_4\) ATPase in fishes and it is related to the transport of Mg\(^2+\) across the gill epithelium (Isaia and Masoni, 1976). This enzyme is also essential for the integrity of the cellular membrane, intracellular cements and for the stabilization of branchial permeability (Potts and Fleming, 1971, Isaia and Masoni, 1976). Na\(^+\) - K\(^+\) ATPase is a membrane bound sulfhydryl containing enzyme whose function is critical for the maintenance of cell viability (Sayeed et al., 2000; Ozcan Oruc et al., 2002). Na\(^+\) - K\(^+\) ATPase is a biochemical expression of active transport of Na\(^+\) and K\(^+\) in the cells (Skou, 1961). This enzyme carries out the transport of sodium and potassium ions against concentration gradient, resulting in the translocation of net charge. The enzyme acts as a current generator and contributes to the membrane potential of the nerve cells (Vizi and Oberfrank,
This enzyme is known to be an early target for oxygen radical induced damage to intact cell (Kim and Akera, 1978; Kako et al., 1988). It is an energy dependent enzyme, which maintains ionic gradients crucial to metabolite transport and osmotic gradients required for the maintenance of cell volume. The transport of Na\(^+\) and K\(^+\) is vital for a number of cellular processes such as maintenance of electrophysicochemical gradients across the cell membranes, especially in nerve and muscle cells (Thomas, 1972), transport of nutrients into interstitial cells (Crane, 1987) and uptake of neurotransmitters in the brain (Iverson and Kelly, 1975).

Osmotic concentration is ultimately bound to the regulation of ionic concentration as well as cell and body volumes (Holmes and Donaldson, 1969; Utida and Hirano, 1973). The uptake of sodium is associated with Na\(^+\) - K\(^+\) ATPase and these numerous studies showing the involvement of this enzyme with the active transport of sodium in fish tissue (Girard and Payan, 1980). Na\(^+\) - K\(^+\) ATPase for example, increases in fish when transferred from freshwater to seawater (Zayugg, 1972; Pfeiler and Kirschner, 1972; Watson 1978). Na\(^+\) - K\(^+\) ATPase displays interdependence with the transport of Na\(^+\) and K\(^+\) which increase presumably to excrete excess salt that has diffused into the fish through the gills (Maetz, 1971; Evans and Mallery, 1975). ATPases are exclusively located in the plasma membrane. Lipid, a major component of cell membranes regulating the activity of membrane ATPases is sensitive to lipophilic inhibitors like pyrethroids and organochlorines (Cutkomp et al., 1982). This suggests that pyrethroid
insecticides decrease ATPase activity by forming an inhibitory complex with the lipid component of the cell membrane.

There are a few reports available on the possible effects of pesticides on the levels of sodium, potassium and calcium ions and the respective ATPase in freshwater animals. Failure of osmoregulatory mechanism have been reported in fish, exposed to pesticides (Grant and Mehrle, 1973). Elevation in serum electrolyte levels has been recorded in some fishes after pesticidal treatment (Eisler and Edmund, 1966; Campbell et al., 1974; Bansal et al., 1979). The flow of ions from exterior to interior or vice versa looks very simple but it is complex electrochemical gradient process. The freshwater organism tend to maintain; their metabolic stability especially through in and out flow of ions. Malla Reddy and Philip (1992) reported significant decrease of whole animal and tissue respiration and also ionic content of sodium, potassium and calcium in the freshwater fish Cyprinus carpio on exposure to fenvalerate.

The permeability of ions is maintained by energy dependent Na\(^+\) - K\(^+\) ATPase system. Few reports are available on pesticide induced alterations in Na\(^+\)-K\(^+\) and Mg\(^{2+}\)ATPase activities (Dalela et al., 1978). Similar observations were also recorded by Janiki and Kunter (1971); Campbell et al., (1974) and Nagendar Reddy (1991). Bansal and Chandra (1985) reported that the insecticide Chlordecon and Mirex inhibits the ATPase activities. Several pesticides have been demonstrated to be inhibitors of ATPase (Desaiah et al., 1980; Bansal and Desaiah, 1982). It has been suggested that mitochondrial (Desaiah and Koch,
1975) and plasma membrane (Matsumura and Narahasi, 1971) ATPase are the targets for their toxic actions. Matsumura (1975) recorded that the pyrethroids (Cypermethrin and Decaroethrin) found to inhibit the ATPases activity in the Squid, Laligo pealei and Malla Reddy et al., (1991) stated that the fenvalerate inhibit the ATPase activity in selected tissues of fish, Cyprinus carpio.

The above cited literature gives an understanding on the effects of pesticides on ionic compostion and associated ATPase activities of freshwater animals. In the present study, an attempt has been made to observe changes involved in the levels of sodium, potassium and calcium ions and Na⁺- K⁺, Mg²⁺ and Ca²⁺ ATPase activities in a few organs of the freshwater fish, Cirrhinus mrigala on short-term and long-term exposure to the lethal and sublethal concentration of fenvalerate.

RESULTS:

Levels of sodium, potassium and calcium ions (µM/ g wet wt) and activities of Na⁺ - K⁺ ATPase, Ca²⁺ ATPase and Mg²⁺ ATPase (µM P/ mg protein/ h) in the three target tissues, namely gill, muscle and liver of freshwater fish, Cirrhinus mrigala on exposure to median lethal concentration of fenvalerate for 1, 2, 3 and 4 days and sub lethal concentration for 1, 5, 10, 15 and 20 days are presented in tables- 41 to 52 and figs. 65 to 76.

SODIUM IONS:

A gradual decrease in the sodium ion level was observed in all the three tissues, namely, gill, muscle and liver under median lethal concentration of
fenvalerate. Maximum mean decrease of -25.176 per cent was observed in gill tissue. Minimum per cent decrease of -2.835 marked the 1st day in liver, while maximum per cent decrease of -45.707 was witnessed on 4th day of exposure in gills (table-41, fig.65).

Sub lethal concentration recorded a continuous decrease in the sodium ion concentration in all the tissues up to day 10. Day 15 and day 20 registered elevation in the sodium ion concentration. Sodium ion levels on day 1 and day 20 in liver showed almost equal rate of decrease, -6.182% and -6.449% respectively. While the values of gill and muscle on day 1 and day 20 differed with very less percent decrease. Liver recorded the maximum mean percent decrease (-11.946) followed by gill (-10.159%) and muscle (-5.812%). Data presented in table-42 and fig. 66 shows values of per cent decrease with every less difference between each tissue and exposure periods.

POTASSIUM IONS :

Potassium ion levels (table-43 & fig.67) showed gradual decrease from day 1 to day 4 in all the three tissues under median lethal concentration. Among the three tissues gill exhibited maximum mean decrease of -20.851 per cent, followed by liver (17.151%) and muscle (-16.650%). Lowest decrease value was noted in liver on day 1 and highest decrease value was recorded in muscle on 4th day of exposure.

Under sublethal concentration, potassium ion concentration decreased continuously up to day 10 in all the three target tissues. While on day 15 and day
20 potassium ion level showed elevated values when compared with the data obtained on day 10. Maximum mean decrease of -13.788 per cent was recorded in liver followed by muscle (-9.232%) and gill (-8.480%). Muscle on first day of exposure to sub lethal concentration showed the least per cent decrease of potassium ion level followed by liver and gill. Maximum per cent decrease of -26.386 per cent was recorded in liver tissue preceded by -18.565 per cent in muscle tissue and -16.307 per cent in gill tissue on 10th day of exposure (table-44 and fig.68).

**CALCIUM IONS:**

Similar tendency as observed in sodium ions and potassium ion levels was observed in the calcium ion levels which also showed gradual decrease in the ionic concentration from day 1 to day 4 on exposure to median lethal concentration of fenvalerate. Maximum per cent decrease was observed in liver (-5.053%) followed by muscle (-6.490%) and gill (-10.828%) on day 1. Gill on 4th day recorded maximum per cent decrease of -38.832 per cent followed by liver (-33.351%) and muscle (-30.489%). Maximum mean per cent decrease was noted in gill (-20.401%) followed by -15.046 per cent in liver and -14.764 per cent in muscle (table-45 and fig.69).

The sub lethal dosage affected a depletion of maximum 29.415 per cent of calcium ions in the gills. In all the tissues from day 1 to day 10 a gradual decrease in the calcium ion level was noted. The day 15 and day 20 showed slight increase in the ion concentration in all the three tissues ranging from 4 to 14 per cent.
Maximum mean per cent decrease was noted in gill (-16.368%) followed by liver (10.374%) and muscle (-8.223%) (table-46 and fig.70).

ATPases:

The activities of Na⁺ - K⁺ ATPase, Ca²⁺ ATPase and Mg²⁺ ATPase are presented in table- 47 to 52 and fig.71 to 76. Na⁺ - K⁺ ATPase activity showed a concurrence with that of the ionic strength of Na⁺ - K⁺, which exhibited a decrease value in the median lethal concentration. Fluctuations in the activity were observed in sub lethal concentration up to day 10 and finally day 15 and day 20 showed elevation in all the three target tissues (gill, muscle and liver). Ca²⁺ ATPase activity showed a gradual decrease at median lethal concentration and variations at sub concentration up to day 10, and enhancement on day 15 and day 20 in gill, muscle and liver. Mg²⁺ ATPase did not differ in exhibiting the trend of decrease in both median lethal and sub lethal concentrations, simultaneously showing an increase on day 15 and day 20 of sub lethal concentration, which was observed in Na⁺ - K⁺ ATPase and Ca²⁺ ATPase.

DISCUSSION:

Strict ion regulation is necessary for aquatic organisms if they are to maintain water and ion homeostasis. The osmotic and ionic characteristics of the body fluids and tissues of freshwater organisms are largely influenced by the ambient medium. The maintenance of homeostasis in such a condition is very much dependent on the osmoregulatory properties and it is a vital phenomenon to
maintain the physiological balance between the external environment and internal milieu of an animal.

Alterations in osmotic regulatory mechanism under toxic stress condition may cause severe imbalance in biochemical composition of the tissue fluids followed by undesirable metabolic consequences. The principal components of osmoregulation are ions. Ions may be anionic or cationic in nature, based on the charge and these help in maintenance of perfect osmolarity of the cell. Osmo and iono-regulation plays an important role in the cellular metabolism of an animal, and its imbalance leads to change in various physiological and bio-chemical activities (Moorthy et al., 1984). Freshwater fishes take up salts from their ambient medium to compensate the water loss through excretion. This obviously necessitates a high metabolic demand. The regulation between the energy and osmoregulation in aquatic animals is well documented by Potts and Parry (1964). Sodium, potassium and calcium are not only important for the maintenance of osmoregulation of body fluids (Baskin et al., 1981), but also for the transport of nutrients from the lumen of the digestive tract into interstitial cells (Crane, 1987) and the uptake of neurotransmitters in the brain (Iverson and Kelly, 1975) . ATPase enzyme complex helps in the uptake of ions from the external medium to the interior of the body of freshwater fishes.

Reports are available to indicate that the insecticides interfere in the transport processes in the biological membrane (Narahashi and Hars, 1968; Rafath
In fishes, gills form a major site for the ion-transport and osmotic water movements, hence, also of pesticides entry. They are the first organs to be exposed to pesticides as they are in constant touch with the polluted water. This affects the permeability characteristics and osmoregulatory function of the gills thereby resulting in the decrease of these ions in gill tissue upon exposure to fenvalerate. In the present study the decrease in the levels of Na\(^+\), K\(^+\) and Ca\(^{2+}\) ions in the gill, muscle and liver exposed to lethal and sublethal concentrations of fenvalerate indicates changes in the permeable properties of the cell membrane of these organs and of deranged Na\(^+\), K\(^+\) and Ca\(^{2+}\) ionic pumps due to the probable consequences of tissues damage. Findings of Kabeer Ahmed et al. (1981) report a decrease in Na\(^+\), K\(^+\) and Ca\(^{2+}\) ions in tissues of fish *Tilapia mossambica* during malathion stress. Moorthy *et al.*, (1984); Malla Reddy and Philip (1991); Siddique *et al.* (1993) offer a strong support for the present observations.

An appraisal of results in the present study suggests that the sodium content decreased as a function of time of exposure to fenvalerate. It is known that sodium content in tissues mainly depend on the permeability functional efficiency of bio-membrane and efficient functional role of Na\(^+\) pump which regulates ionic content of tissues. The level of Na\(^+\) signifies the tissue importance in the mobilization of water transport, since sodium content in the membrane facilitates the water movement among the tissue (Wilbur, 1972; Dietz, 1979). From the
result, this is evident that the Na\(^+\) loss is higher in the case of gill indicating the derangement in Na\(^+\) transport. Also, the decreased sodium content in the tissues of exposed fish indicates changes in permeable properties of different bio-membrane systems to different extent by altering the Na\(^+\) pump and rupture in the respiratory epithelium of gill tissue (Radhaiah, 1988).

On exposure to fenvalerate, Ca\(^{2+}\) was decreased indicating increased decalcification. It is known that Ca\(^{2+}\) is concerned with neuromuscular excitability, cell membrane permeability and regulation of protein binding capacity (Walser, 1960). In the present study, the restlessness in *Cirrhinus mrigala* during pyrethroid stress might indicate alterations in the regulations of Ca\(^{2+}\) in the tissue. Moreover, it has been reported that decreased calcium content during pesticide stress corresponds to structural changes in mitochondria integrity (Miroslaw, 1973). Since mitochondria acts as "Sinks" for intra cellular Ca\(^{2+}\) (Bygrave, 1978) and principal store houses of Ca\(^{2+}\) deposition, it appears that the decreased Ca\(^{2+}\) in the present study might attribute to the disturbances in mitochondrial integrity and subsequent respiratory distress. Hoar (1976) suggested that the levels of amino acids and metabolites like pyruvate and lactate will be increased under stress conditions to compensate the loss of inorganic ions. Amino acids and lactate were found increased in the tissues of *Cyprinus carpio* and *Labeo rohita* exposed to sublethal concentration of fenvalerate (Malla Reddy *et al.*, 1991) and cypermethrin (Sridevi, 1991) causing metabolic diversion in fish to prolong its survivability under severe osmotic imbalance.
It is known that any remarkable decrease in K⁺ level might be accompanied by serious disturbances in muscular irritability, myocardial function and respiration (Coles, 1967). The decrease in K⁺ content in the tissues of Cirrhinus mrigala exposed to fenvalerate might be attributed to the derangement in respiration at whole animal as observed in the present investigation.

The main reason for the decrease in sodium, potassium and calcium ion levels in the organs of fish, exposed to fenvalerate could be attributed to the suppressed activities of Na⁺ - K⁺ Mg²⁺ and Ca²⁺ ATPase (Renfro et al., 1974). Since ATPases have been described as prominent energy linked enzymes in fishes (Desaiah et al., 1975), inhibition of these enzymes by fenvalerate influences the movement of ions by active transport. The suppression in ATPase activities also suggests a drastic decrease in the prolactin release, which might be particularly responsible for the hypocalcemia. Rapid induction of hypocalcemia by cadmium has been reported in rainbow trout (Roch and Maly 1979; Giles, 1984), Flounder (Larsson et al., 1981), Carp (Koyama and Itazawa, 1977; Yamawaki et al., 1986) and Tilapia (Pratap et al., 1989).

It is quite evident that the fish, Cirrhinus mrigala under fenvalerate stress seldom undergoes total abolition of functional regulation of the ionic transport and water permeability and the imbalance in osmoregulation is compensated in hormonic fashion through the production of biochemically changed components like amino acids which go to rescue and compensate the imbalance ionic composition. Thus, inherent osmoregulation of freshwater fish is viewed in a
nutshell but clear understanding on the basis of water permeability versus Na' transport and the role of Ca\textsuperscript{2+} in the water transport mechanism still awaits direct analyses.

The activities of Mg\textsuperscript{2+}, Na\textsuperscript{+} and Ca\textsuperscript{2+} ATPases decreased in gill, muscle and liver of the fish on exposure to sublethal concentrations of fenvalerate. The decrease in these activities indicates the demolition of cellular ionic regulation in the organs of the fish as reported by Renfro et al., (1974) and Schmidt Nielson (1975). This disruption may be due to the effect of fenvalerate (pyrethroid) on passive movement of ions i.e., the permeability characteristics. In this connection, it is of interest to note that \textit{O\textsubscript{2}} consumption was decreased in the fish \textit{Cyprinus carpio} under fenvalerate and cypermethrin stress (Malla Reddy, 1987) and in \textit{Cirrhinus mrigala} (Mushigeri and David, 2003), \textit{Labeo rohita} (David et al., 2002) exposed to fenvalerate and also on oxidative metabolism as measured by the activities of ICDH, SDH, MDH and cytochrome-c-oxidase which have been inhibited in the freshwater fish, \textit{Labeo rohita} exposed to cypermethrin (Ghosh, 1989) and in the prawn, \textit{Metapenaeus monoceros} exposed to fenvalerate (Malla Reddy et al., 1987). The decrease in activities may also be due to interaction of pesticide with Mg\textsuperscript{2+} and Na\textsuperscript{+} - K\textsuperscript{+} ATPases thereby inducing inhibition (Dikshith et al., 1978).

ATPase, a membrane bound enzyme group, are responsible for the movement of different ions across the membrane. In fish, various toxicants and ions enter into the body by absorption and adsorption by the gill surface and then
followed by diffusion. An interaction with the membrane may disrupt the osmotic and ionic regulation of the gill tissue by affecting the membrane permeability, mainly due to inactivation of the ATPase in the bronchial epithelial cells. This ATPase system is active in all parts of the intestine, but has maximum activity in the hind end of the intestinal tract, so that a major portion of the digested nutrients, salts followed by water are absorbed by hind gut. The ion dependent ATPases are known to regulate the entry and exit of different ions across the membrane, in order to maintain the physiological requirements of the cell (Jignasa et al., 1997).

It is known that the pyrethroid causes the disturbances in neural transmission (Bandhopadyay, 1982; Yellamma and Reddy, 1987) and such a disturbance might lead to perturbations in ATPase system. Portella et al., (1963) have suggested that the decrease in Mg$^{2+}$ ATPase activity might be due to the damage of the mitochondrial membranes which may interfere with the conversion of oxidative energy to phosphate bond energy. Decrease in Ca$^{2+}$, Mg$^{2+}$ ATPase activity in Loligo pealei exposed to allethrin, permethrin and cypermethrin reported by Clark and Matsumura (1982) offers best support to the present study. Since Mg$^{2+}$ ATPase is involved in oxidative phosphorylation (Durairaj, 2001), the inhibition due to fenvalerate toxicity directly prevents or reduces the oxidative phosphorylation. Neil (1968) has reported that uncoupling agents increase the hydrolysis of ATP and inhibits the phosphorylation. This mechanism may be operating in the pesticide treated fish and impair the energy producing system.
Since this enzyme is directly involved in the oxidative phosphorylation the action of pesticide on this system correlates with toxicity of pesticide.

According to Price (1978) the inhibition is due to phosphorylation of active site of the enzyme as in the case of acetylcholinesterase inhibition. Since Na⁺ - K⁺ ATPase is considered as a marker enzyme to understand the physiological impairment of the cell (Campbell et al., 1974). The inhibition reveals the disruption of ionic movement in neuronal and glial cells. Such alterations in ionic balance depolarize the nerve and due to depolarization the nerve cells increase in the releasing of neurotransmitter (Kimelberg and Papahad Jopulos, 1974) which in turn inhibits Na⁺ - K⁺ ATPase activity (Stojanovic et al., 1980). Similar results and conclusion was drawn by Durairaj (2001).

Prasada Rao et al., (1984), reported that the activities of OS-Mg²⁺ and Na⁺ - K⁺ ATPases were decreased in rat exposed to pyrethroids, which supports the findings of the present study, and the ATPase levels reduced in Daphnia magna exposed to fenvalerate (McKee and knowles, 1986). Na⁺- K⁺ and Ca²⁺ - Mg²⁺ ATPase activities were inhibited in the squid Laligo pealei by the two types of pyrethroids (Clark and Matsumura, 1982).

The inhibition of ATPase activities in the present study and greater decrease in the levels of ions observed in the gill, muscle and liver of fish, exposed to the lethal concentrations of fenvalerate indicate the effects of fenvalerate on osmo-ionoregulations of this animal. As the ion regulatory capacity is energy dependent process the greater decrease in the energy releasing pathways
in fish subjected to lethal intoxication provides support for the more decrease in the levels of Na\(^+\), K\(^+\) and Ca\(^{2+}\) ions. Further greater imbalance caused to the gill structures could also be one of the probable reasons for observed perturbations of ATPase activities and ionic levels in the fish. At cellular level the availability of pesticide to interact with the ATPase might depend on the cell surface area. The inhibition of Na\(^+\) - K\(^+\) ATPases probably disturbed Na\(^+\) - K\(^+\) pump, resulting in an uncontrollable entry of Na\(^+\) into the cell along the concentration gradient. This process may cause swelling of the cell and finally membrane ruptures. In teleosts Na\(^+\) - K\(^+\) ATPases transport Na\(^+\) and Cl\(^-\) by gills. It has been reported that the high activity of Na\(^+\) - K\(^+\) ATPases in marine teleosts cause intense ionic fluxes and low branchial osmotic permeability (Sargent et al., 1980). Inhibition of this enzyme by the toxicant, fenvalerate presumably prevents the build up of high ion concentration in the extracellular spaces resulting in block of the movement of ions towards the external medium via the leakage junctions (Kundu et al., 1986).

However, in the sublethal concentrations significant elevation in ion levels and also in the ATPase activities in the organs of fish, from 15 days of exposure to 20 days of exposure, indicate the greater efficiency to resist the sublethal concentration of fenvalerate. Recovery of ATPase in the freshwater crab *Oziotelphusa senex senex* exposed to sublethal concentration of endosulfan was reported by Malla Reddy et al., (1991). This could be due to their higher protein synthetic ability. The increase in oxidative metabolism also might have facilitated these animals to elevate the ionic strength by meeting the energy demands. The increase in the ionic concentration may be helpful to the fish for the maintenance
of higher osmotic gradient in order to curb the speedy entry of toxicant. Further the increase in ion levels may elevate the neuromuscular activity for the enhancement in their synthetic potentials particularly related to pesticide detoxification and domination process. Also the increased ions may help the easy uptake of the metabolites and the structural rigidity in cellular construction. The ability to recover from the state of imbalance was seen at 15 days of exposure, but maintained at 20 days with an initial struggle for survival.

In support to the findings of the present study, it has been reported that Na⁺ - K⁺ ATPase activity in liver of *C. gachua* exposed to endosulfan was significantly decreased (Sharma, 1988). Exposure to fenithrothion decreased ATPase activity of *Anguilla anguilla* (Sancho *et al.*, 1997). Na⁺ - K⁺ ATPase activity was decreased in rats exposed to a new phosphorthionate (RPR-II) (Rahaman *et al.*, 2000). Ozcan Oruc *et al.*, (2002) reported increased lipid peroxide formation in three fish species exposed to azinphosmethyl, which could disturb the anatomical integrity of the biomembrane and diminish its fluidity leading to inhibition of several membrane bound enzymes including Na⁺ - K⁺ ATPase. It is reported that pyrethroid, cypermethrin; mexacrabate and phorate impair the stability of the cell membrane by damaging its structural lipid by peroxidation decomposition, which may lead to subsequent cell necrosis and functional dearrangement (Sing *et al.*, 1993). Shaheen *et al.*, (1996) suggested that free radicals generated by these disorders attack the membrane phospholipids, causing their peroxidation. These peroxidative processes are surely contributing to
the inactivation of membrane bound biomolecules such as enzymes, since phospholipids are important for the optimum activity of many enzymes.

Greater degree of decrease in Na⁺, K⁺ and Ca²⁺ levels and the activities of Na⁺- K⁺, Mg²⁺ and Ca²⁺ ATPase in the fish exposed to the lethal concentration of fenvalerate indicates severe disruption in the cellular ionic regulation. High concentration of fenvalerate might have greatly altered the permeability characteristics of the membranes of the organs by interacting with the membrane proteins readily to serve alterations in the acute transport through destabilizing the membrane bound enzymes and related hormonal and energy producing process. Further, the progressive decrease in the ion levels and progressive suppression of Na⁺- K⁺, Mg²⁺ and Ca²⁺ ATPase activities in the organs of fish, over time of exposure to the lethal concentrations of fenvalerate indicate the increase in the binding of the fenvalerate to the active sites of membrane bound enzymes as the degree of inhibition is dependent on the concentration of fenvalerate available to the active sites on enzyme molecules. The drastic decrease in the rate of oxygen consumption and oxidative metabolic cycles in the organs of fish, from day 1 to day 4 in the lethal concentration also lend support for the study of decrease in the Na⁺, K⁺ and Ca²⁺- ionic levels and concentrations of associated ATPase activities.

In sublethal concentration of fenvalerate the Na⁺- K⁺ and Ca²⁺ levels significantly decreased with ion-competent inhibition of associated Na⁺- K⁺, Ca²⁺ and Mg²⁺ ATPase activities in the organs of fish at 1 day and 10 day exposures. The significant elevation in the ionic levels and enzyme activities at 15 and 20
days indicate that on prolonged exposure the sub acute concentration of fenvalerate could not elicit inhibitory effect either on the uptake of ions or on the activities of ATPase and instead it stimulated the uptake. Possibly the inhibition of ATPase activity is dependent on the functional groups of the enzyme and the amount of fenvalerate available for the competitive replacement of the substrate. Further recruitment of chloride cells has been proposed as a fundamental and physiologically significant response of fresh water fish to increase the capability to take up Na⁺ K⁺ and Ca²⁺ from water (Leino et al., 1987, Fu et al., 1990). Even the secretion might be increased to induce particularly hypercalcemia by remobilization of Ca²⁺ from exchangeable Ca²⁺ stores. Muramotao (1981) reported the direct uptake via gills. All these factors could be an active operation for the elevation in ion levels and ATPase activities in the gills, muscle and liver of fish from 15 to 20 days. The increased ionic level may be helpful to the animal to prevent the entry of toxic fenvalerate by maintaining cation concentration gradient.
Table 41

Sodium ion content (μM/g wet wt) in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Exposure Periods</th>
<th>Mean</th>
<th>S.Em±</th>
<th>C. V. (%)</th>
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<tr>
<td></td>
<td></td>
<td>Day 1</td>
<td>Day 2</td>
<td>Day 3</td>
<td>Day 4</td>
</tr>
<tr>
<td>GILL</td>
<td>51.532 a</td>
<td>43.738 b</td>
<td>39.438 c</td>
<td>30.101 d</td>
<td>27.978 e</td>
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<tr>
<td></td>
<td>0.00019</td>
<td>0.00023</td>
<td>0.00028</td>
<td>0.00023</td>
<td>0.00027</td>
</tr>
<tr>
<td>MUSCLE</td>
<td>39.384 a</td>
<td>34.684 b</td>
<td>29.904 c</td>
<td>23.060 d</td>
<td>21.715 e</td>
</tr>
<tr>
<td></td>
<td>0.00027</td>
<td>0.00019</td>
<td>0.00031</td>
<td>0.00023</td>
<td>0.00026</td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td>-11.933</td>
<td>-24.070</td>
<td>-41.448</td>
<td>-44.863</td>
</tr>
<tr>
<td>LIVER</td>
<td>47.683 a</td>
<td>46.332 b</td>
<td>41.069 c</td>
<td>36.599 d</td>
<td>32.869 e</td>
</tr>
<tr>
<td></td>
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<td>0.00028</td>
<td>0.00028</td>
<td>0.01560</td>
<td>0.00022</td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td>-2.833</td>
<td>-13.870</td>
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</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p<0.001) from each other according to Duncan’s Multiple Range (DMR) Test.

SD± : Standard Deviation
S.Em± : Standard error of means.
C. V : Critical Value
Table-42

Sodium ion content (μM/g wet wt) in the organs of the fish, *Cirrhinus mrigala* on exposure to sublethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Exposure Periods</th>
<th>Mean</th>
<th>S.Em±</th>
<th>C.V. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Day 1</td>
<td>Day 5</td>
<td>Day 10</td>
<td>Day 15</td>
</tr>
<tr>
<td>GILL</td>
<td>51.532 a</td>
<td>47.484 c</td>
<td>44.442 e</td>
<td>40.939 f</td>
<td>45.372 d</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00019</td>
<td>0.00022</td>
<td>0.00028</td>
<td>0.00028</td>
<td>0.00035</td>
</tr>
<tr>
<td>MUSCLE</td>
<td>39.384 a</td>
<td>36.836 a</td>
<td>33.728 a</td>
<td>30.928 a</td>
<td>34.682 a</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00027</td>
<td>0.00026</td>
<td>0.00244</td>
<td>0.00257</td>
<td>0.00024</td>
</tr>
<tr>
<td>LIVER</td>
<td>47.683 a</td>
<td>44.735 b</td>
<td>39.910 d</td>
<td>36.387 f</td>
<td>38.624 e</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00024</td>
<td>0.00024</td>
<td>0.00031</td>
<td>0.00029</td>
<td>0.00028</td>
</tr>
<tr>
<td>% change</td>
<td>--------</td>
<td>-6.182</td>
<td>-16.301</td>
<td>-23.690</td>
<td>-18.999</td>
</tr>
</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p<0.001) from each other according to Duncan's Multiple Range (DMR) Test.

SD± : Standard Deviation
S.Em± : Standard error of means.
C.V : Critical Value
Fig. 65: Per cent decrease over control in Na⁺ ion content in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate

Fig. 66: Per cent decrease over control in Na⁺ ion content in the organs of the fish, *Cirrhinus mrigala* on exposure to sub lethal concentration of fenvalerate
Table 43

Potassium ion content (µM/g wet wt) in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Exposure Periods</th>
<th>Mean</th>
<th>S.Em±</th>
<th>C.V. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Day 1</td>
<td>Day 2</td>
<td>Day 3</td>
<td>Day 4</td>
</tr>
<tr>
<td>GILL</td>
<td>59.396 a</td>
<td>50.064 b</td>
<td>47.862 c</td>
<td>41.729 d</td>
<td>36.002 e</td>
</tr>
<tr>
<td>SD±</td>
<td>0.00116</td>
<td>0.00023</td>
<td>0.00024</td>
<td>0.00292</td>
<td>0.00027</td>
</tr>
<tr>
<td>MUSCLE</td>
<td>67.600 a</td>
<td>61.001 b</td>
<td>56.346 c</td>
<td>50.407 d</td>
<td>46.364 e</td>
</tr>
<tr>
<td>SD±</td>
<td>0.00022</td>
<td>0.00024</td>
<td>0.00024</td>
<td>0.00022</td>
<td>0.00026</td>
</tr>
<tr>
<td>% change</td>
<td>--------</td>
<td>-9.761</td>
<td>-16.647</td>
<td>-25.433</td>
<td>-44.613</td>
</tr>
<tr>
<td>LIVER</td>
<td>51.956 a</td>
<td>47.999 b</td>
<td>41.817 c</td>
<td>38.428 d</td>
<td>35.026 e</td>
</tr>
<tr>
<td>SD±</td>
<td>0.00023</td>
<td>0.00019</td>
<td>0.00019</td>
<td>0.00022</td>
<td>0.00019</td>
</tr>
<tr>
<td>% change</td>
<td>--------</td>
<td>-7.616</td>
<td>-19.514</td>
<td>-26.037</td>
<td>-32.585</td>
</tr>
</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p≤0.001) from each other according to Duncan's Multiple Range (DMR) Test.

SD± : Standard Deviation
S.Em± : Standard error of means.
C.V : Critical Value
Potassium ion content (µM /g wet wt) in the organs of the fish, *Cirrhinus mrigala* on exposure to sublethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Exposure Periods</th>
<th></th>
<th></th>
<th></th>
<th>Mean</th>
<th>S.Em±</th>
<th>C.V. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Day 1</td>
<td>Day 5</td>
<td>Day 10</td>
<td>Day 15</td>
<td>Day 20</td>
<td></td>
<td></td>
</tr>
<tr>
<td>GILL</td>
<td>59.396 a</td>
<td>56.096 b</td>
<td>52.748 d</td>
<td>49.710 f</td>
<td>52.374 e</td>
<td>55.831 c</td>
<td>54.359</td>
<td>0.0002</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00116</td>
<td>0.00022</td>
<td>0.00019</td>
<td>0.00019</td>
<td>0.00024</td>
<td>0.00019</td>
<td></td>
<td></td>
</tr>
<tr>
<td>MUSCLE</td>
<td>67.600 a</td>
<td>64.474 b</td>
<td>59.749 d</td>
<td>55.050 f</td>
<td>59.076 e</td>
<td>62.203 c</td>
<td>61.359</td>
<td>0.0001</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00022</td>
<td>0.00023</td>
<td>0.00024</td>
<td>0.00033</td>
<td>0.00024</td>
<td>0.00019</td>
<td></td>
<td></td>
</tr>
<tr>
<td>LIVER</td>
<td>51.956 a</td>
<td>48.458 b</td>
<td>43.006 d</td>
<td>38.002 f</td>
<td>41.364 e</td>
<td>45.964 c</td>
<td>44.792</td>
<td>0.0306</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00023</td>
<td>0.00019</td>
<td>0.00023</td>
<td>0.00023</td>
<td>0.00023</td>
<td>0.00023</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p<0.001) from each other according to Duncan’s Multiple Range (DMR) Test.
SD± : Standard Deviation
S.Em± : Standard error of means.
C.V : Critical Value
Fig. 67: Per cent decrease over control in K⁺ ion content in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate.

Fig. 68: Per cent decrease over control in K⁺ ion content in the organs of the fish, *Cirrhinus mrigala* on exposure to sub lethal concentration of fenvalerate.
Table-45

Calcium ion content (µM/g wet wt) in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Mean</th>
<th>S.Em±</th>
<th>C.V. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Exposure Periods</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Day 1</td>
<td>Day 2</td>
<td>Day 3</td>
</tr>
<tr>
<td><strong>GILL</strong></td>
<td>58.992 a</td>
<td>52.604 b</td>
<td>46.721 c</td>
<td>40.384 d</td>
</tr>
<tr>
<td><strong>SD±</strong></td>
<td>0.00019</td>
<td>0.00019</td>
<td>0.00024</td>
<td>0.00210</td>
</tr>
<tr>
<td>% change</td>
<td>-------</td>
<td>-10.828</td>
<td>-20.801</td>
<td>-31.543</td>
</tr>
<tr>
<td><strong>MUSCLE</strong></td>
<td>71.687 a</td>
<td>67.034 b</td>
<td>61.473 c</td>
<td>55.492 d</td>
</tr>
<tr>
<td><strong>SD±</strong></td>
<td>0.00019</td>
<td>0.00022</td>
<td>0.00019</td>
<td>0.00023</td>
</tr>
<tr>
<td><strong>LIVER</strong></td>
<td>64.073 a</td>
<td>60.835 b</td>
<td>54.638 c</td>
<td>49.908 d</td>
</tr>
<tr>
<td><strong>SD±</strong></td>
<td>0.00029</td>
<td>0.00019</td>
<td>0.00019</td>
<td>0.00023</td>
</tr>
<tr>
<td>% change</td>
<td>-------</td>
<td>-5.053</td>
<td>-14.725</td>
<td>-22.107</td>
</tr>
</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p≤0.001) from each other according to Duncan's Multiple Range (DMR) Test.

**SD±** : Standard Deviation

**S.Em±** : Standard error of means.

**C.V**  : Critical Value
Table 46

Calcium ion content (μM/g wet wt) in the organs of the fish, *Cirrhinus mrigala* on exposure to sublethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Exposure Periods</th>
<th>Mean</th>
<th>S.Em±</th>
<th>C.V (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Day 1</td>
<td>Day 5</td>
<td>Day 10</td>
<td>Day 15</td>
</tr>
<tr>
<td>GILL</td>
<td>58.992</td>
<td>a</td>
<td>53.750</td>
<td>b</td>
<td>47.839</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00019</td>
<td></td>
<td>0.00023</td>
<td>0.00019</td>
<td>0.00019</td>
</tr>
<tr>
<td>MUSCLE</td>
<td>71.687</td>
<td>a</td>
<td>67.503</td>
<td>c</td>
<td>63.382</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00019</td>
<td></td>
<td>0.00033</td>
<td>0.00055</td>
<td>0.00025</td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td></td>
<td>-5.836</td>
<td>-11.585</td>
<td>-17.330</td>
</tr>
<tr>
<td>LIVER</td>
<td>64.073</td>
<td>a</td>
<td>61.004</td>
<td>b</td>
<td>56.655</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00029</td>
<td></td>
<td>0.00019</td>
<td>0.00019</td>
<td>0.00026</td>
</tr>
</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p<0.001) from each other according to Duncan's Multiple Range (DMR) Test.

SD± : Standard Deviation
S.Em± : Standard error of means.
C.V : Critical Value
Exposure period in days

Per cent decrease over control in Ca²⁺ ion content in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate.

Fig. 69: Per cent decrease over control in Ca²⁺ ion content in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate.

Exposure period in days

Per cent decrease over control in Ca²⁺ ion content in the organs of the fish, *Cirrhinus mrigala* on exposure to sub lethal concentration of fenvalerate.

Fig. 70: Per cent decrease over control in Ca²⁺ ion content in the organs of the fish, *Cirrhinus mrigala* on exposure to sub lethal concentration of fenvalerate.
Table-47

Na\(^+\) - K\(^+\) ATPase activity (\(\mu\)M of Pi formed /mg protein /h) in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Exposure Periods</th>
<th>Mean</th>
<th>S.Em±</th>
<th>C.V. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Day 1</td>
<td>Day 2</td>
<td>Day 3</td>
<td>Day 4</td>
</tr>
<tr>
<td>GILL</td>
<td>8.920 a</td>
<td>8.066 b</td>
<td>7.109 c</td>
<td>6.021 d</td>
<td>4.998 e</td>
</tr>
<tr>
<td></td>
<td>0.00026</td>
<td>0.00025</td>
<td>0.00030</td>
<td>0.00030</td>
<td>0.00031</td>
</tr>
<tr>
<td>MUSCLE</td>
<td>6.734 a</td>
<td>6.439 b</td>
<td>5.976 c</td>
<td>5.061 d</td>
<td>4.653 e</td>
</tr>
<tr>
<td></td>
<td>0.00031</td>
<td>0.00030</td>
<td>0.00022</td>
<td>0.00040</td>
<td>0.00033</td>
</tr>
<tr>
<td>LIVER</td>
<td>5.939 a</td>
<td>5.684 b</td>
<td>5.096 c</td>
<td>4.532 d</td>
<td>4.012 e</td>
</tr>
<tr>
<td></td>
<td>0.00024</td>
<td>0.00031</td>
<td>0.00023</td>
<td>0.00030</td>
<td>0.00027</td>
</tr>
<tr>
<td></td>
<td>-----</td>
<td>-4.293</td>
<td>-14.194</td>
<td>-23.690</td>
<td>-32.446</td>
</tr>
</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p<0.001) from each other according to Duncan's Multiple Range (DMR) Test.

SD± : Standard Deviation
S.Em± : Standard error of means.
C.V : Critical Value
Table-48

Na⁺ – K⁺ ATPase activity (µM of Pi formed /mg protein /h) in the organs of the fish, *Cirrhinus mrigala* on exposure to sublethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Exposure Periods</th>
<th>Mean</th>
<th>S.Em±</th>
<th>C.V. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Day 1</td>
<td>Day 5</td>
<td>Day 10</td>
<td>Day 15</td>
</tr>
<tr>
<td>GILL</td>
<td>8.920 a</td>
<td>8.103 b</td>
<td>7.900 d</td>
<td>7.501 e</td>
<td>8.009 c</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00026</td>
<td>0.00029</td>
<td>0.00034</td>
<td>0.00036</td>
<td>0.00025</td>
</tr>
<tr>
<td>MUSCLE</td>
<td>6.734 a</td>
<td>6.539 b</td>
<td>5.332 e</td>
<td>4.984 f</td>
<td>5.990 d</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00031</td>
<td>0.00057</td>
<td>0.00033</td>
<td>0.00031</td>
<td>0.00033</td>
</tr>
<tr>
<td>LIVER</td>
<td>5.939 a</td>
<td>5.830 b</td>
<td>5.120 d</td>
<td>4.743 f</td>
<td>5.024 e</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00024</td>
<td>0.00013</td>
<td>0.00029</td>
<td>0.00028</td>
<td>0.00025</td>
</tr>
</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p<0.001) from each other according to Duncan’s Multiple Range (DMR) Test.

SD± : Standard Deviation
S.Em± : Standard error of means.
C.V : Critical Value
Fig. 71: Per cent decrease over control in Na⁺ - K⁺ ATPase activity in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate.

Fig. 72: Per cent decrease over control in Na⁺ - K⁺ ATPase activity in the organs of the fish, *Cirrhinus mrigala* on exposure to sub lethal concentration of fenvalerate.
Table-49

Ca^{2+} ATPase activity (µM of Pi formed /mg protein /h) in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Exposure Periods</th>
<th>Mean</th>
<th>S.Em±</th>
<th>C.V. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Day 1</td>
<td>Day 2</td>
<td>Day 3</td>
<td>Day 4</td>
</tr>
<tr>
<td>GILL</td>
<td>7.893 a</td>
<td>6.743 b</td>
<td>6.030 c</td>
<td>5.127 d</td>
<td>4.218 e</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00028</td>
<td>0.00033</td>
<td>0.00023</td>
<td>0.00019</td>
<td>0.00029</td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td>-14.569</td>
<td>-23.603</td>
<td>-35.043</td>
<td>-46.560</td>
</tr>
<tr>
<td>MUSCLE</td>
<td>4.819 a</td>
<td>4.139 b</td>
<td>3.246 c</td>
<td>2.448 d</td>
<td>1.601 e</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00033</td>
<td>0.00023</td>
<td>0.00019</td>
<td>0.00028</td>
<td>0.00045</td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td>-14.110</td>
<td>-32.641</td>
<td>-49.201</td>
<td>-66.777</td>
</tr>
<tr>
<td>LIVER</td>
<td>2.383 a</td>
<td>1.839 b</td>
<td>1.268 c</td>
<td>0.854 d</td>
<td>0.391 e</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00043</td>
<td>0.00035</td>
<td>0.00035</td>
<td>0.00035</td>
<td>0.00034</td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td>-22.828</td>
<td>-46.789</td>
<td>-64.162</td>
<td>-83.592</td>
</tr>
</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p≤0.001) from each other according to Duncan's Multiple Range (DMR) Test.

SD= : Standard Deviation
S.Em± : Standard error of means.
C.V : Critical Value
Ca\textsuperscript{2+} ATPase activity (\(\mu\text{M of Pi formed /mg protein /h}\)) in the organs of the fish, *Cirrhinus mrigala* on exposure to sublethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Day 1</th>
<th>Day 5</th>
<th>Day 10</th>
<th>Day 15</th>
<th>Day 20</th>
<th>Mean</th>
<th>S.Em±</th>
<th>C. V. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GILL</td>
<td>7.893 a</td>
<td>6.106 b</td>
<td>5.473 bc</td>
<td>4.204 d</td>
<td>5.188 c</td>
<td>5.236 c</td>
<td>5.683</td>
<td>0.1839</td>
<td>7.92</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00028</td>
<td>0.00036</td>
<td>0.00042</td>
<td>0.00025</td>
<td>0.01074</td>
<td>0.00029</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td>-22.64</td>
<td>-30.660</td>
<td>-46.737</td>
<td>-34.270</td>
<td>-33.662</td>
<td>-27.999</td>
<td></td>
<td></td>
</tr>
<tr>
<td>MUSCLE</td>
<td>4.819 a</td>
<td>4.478 b</td>
<td>3.882 c</td>
<td>2.003 f</td>
<td>2.975 f</td>
<td>3.735 d</td>
<td>3.649</td>
<td>0.0001</td>
<td>0.01</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00033</td>
<td>0.00028</td>
<td>.00034</td>
<td>0.00028</td>
<td>0.00028</td>
<td>0.00043</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LIVER</td>
<td>2.383 a</td>
<td>2.108 b</td>
<td>1.759 c</td>
<td>1.330 e</td>
<td>1.692 d</td>
<td>2.105 b</td>
<td>1.896</td>
<td>0.0001</td>
<td>0.02</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00043</td>
<td>0.00035</td>
<td>0.00034</td>
<td>0.00030</td>
<td>0.00037</td>
<td>0.00031</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p<0.001) from each other according to Duncan’s Multiple Range (DMR) Test.

SD± : Standard Deviation
S.Em± : Standard error of means.
C. V. : Critical Value
Fig. 73: Per cent decrease over control in Ca\textsuperscript{2+} ATPase activity in the organs of the fish, Cirrhinus mrigala on exposure to median lethal concentration of fenvalerate.

Fig. 74: Per cent decrease over control in Ca\textsuperscript{2+} ATPase activity in the organs of the fish, Cirrhinus mrigala on exposure to sub lethal concentration of fenvalerate.
Table 51

Mg²⁺ ATPase activity (μM of Pi formed /mg protein /h) in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Exposure Periods</th>
<th>Mean</th>
<th>S.Em±</th>
<th>C.V. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Day 1</td>
<td>Day 2</td>
<td>Day 3</td>
<td>Day 4</td>
</tr>
<tr>
<td>GILL</td>
<td>7.646 a</td>
<td>6.814 b</td>
<td>6.236 c</td>
<td>5.632 d</td>
<td>4.863 e</td>
</tr>
<tr>
<td>SD±</td>
<td>0.00024</td>
<td>0.00040</td>
<td>0.01215</td>
<td>0.00023</td>
<td>0.00033</td>
</tr>
<tr>
<td>MUSCLE</td>
<td>4.776 a</td>
<td>4.328 b</td>
<td>3.793 c</td>
<td>3.219 d</td>
<td>2.644 e</td>
</tr>
<tr>
<td>SD±</td>
<td>0.00031</td>
<td>0.00029</td>
<td>0.00035</td>
<td>0.00028</td>
<td>0.00047</td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td>-9.380</td>
<td>-20.582</td>
<td>-32.600</td>
<td>-44.639</td>
</tr>
<tr>
<td>LIVER</td>
<td>2.645 a</td>
<td>2.192 b</td>
<td>1.786 c</td>
<td>1.464 d</td>
<td>0.999 e</td>
</tr>
<tr>
<td>SD±</td>
<td>0.00024</td>
<td>0.00039</td>
<td>0.00032</td>
<td>0.00028</td>
<td>0.00036</td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td>-17.126</td>
<td>-32.476</td>
<td>-44.650</td>
<td>-62.230</td>
</tr>
</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p<0.001) from each other according to Duncan's Multiple Range (DMR) Test.

SD= : Standard Deviation
S.Em± : Standard error of means.
C.V. : Critical Value
Table-52

Mg\(^{2+}\) ATPase activity (µM of Pi formed /mg protein/h) in the organs of the fish, *Cirrhinus mrigala* on exposure to sublethal concentration of fenvalerate.

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Control</th>
<th>Exposure Periods</th>
<th>Mean</th>
<th>S.Em±</th>
<th>C.V. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Day 1</td>
<td>Day 5</td>
<td>Day 10</td>
<td>Day 15</td>
</tr>
<tr>
<td>GILL</td>
<td>7.646 a</td>
<td>7.432 b</td>
<td>6.721 c</td>
<td>6.111 e</td>
<td>6.520 d</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00024</td>
<td>0.00024</td>
<td>0.00029</td>
<td>0.00026</td>
<td>0.00035</td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td>-2.798</td>
<td>-12.097</td>
<td>-20.075</td>
<td>-14.726</td>
</tr>
<tr>
<td>MUSCLE</td>
<td>4.776 a</td>
<td>4.538 b</td>
<td>4.002 e</td>
<td>3.663 f</td>
<td>4.202 d</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00031</td>
<td>0.00037</td>
<td>0.00019</td>
<td>0.00054</td>
<td>0.00031</td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td>-4.983</td>
<td>-16.206</td>
<td>-23.307</td>
<td>-12.018</td>
</tr>
<tr>
<td>LIVER</td>
<td>2.645 a</td>
<td>2.508 b</td>
<td>2.013 d</td>
<td>1.786 e</td>
<td>2.007 d</td>
</tr>
<tr>
<td>SD ±</td>
<td>0.00024</td>
<td>0.00033</td>
<td>0.00026</td>
<td>0.00037</td>
<td>0.00036</td>
</tr>
<tr>
<td>% change</td>
<td>------</td>
<td>-5.179</td>
<td>-23.894</td>
<td>-32.476</td>
<td>-24.120</td>
</tr>
</tbody>
</table>

Means are SD± (n=6) for a organ in a row followed by the same letter are not significantly different (p<0.001) from each other according to Duncan's Multiple Range (DMR) Test.

SD± : Standard Deviation
S.Em± : Standard error of means.
C.V : Critical Value
Fig. 75: Per cent decrease over control in Mg$^{2+}$ ATPase activity in the organs of the fish, *Cirrhinus mrigala* on exposure to median lethal concentration of fenvalerate.

Fig. 76: Percent decrease over control in Mg$^{2+}$ ATPase activity in the organs of the fish, *Cirrhinus mrigala* on exposure to sub lethal concentration of fenvalerate.