Discussion
5. DISCUSSION

5.1. Iron epilepsy

The iron-induced experimental epilepsy, like the post-traumatic clinical epilepsy, is associated with an iron-induced acquired lesion. An epileptogenic focus develops around this lesion. The structure of the lesion is of considerable interest. Light microscopic structure of the iron-induced lesion has been described in earlier studies (Willmore and Rubin, 1982; Willmore et al., 1978). In order to ensure that the lesion produced in the present experiments was similar to that reported in published reports, structure of the lesion was studied. Since the light microscopic structure of the iron-induced lesion has already been described in several published reports, electron microscopic structure of the lesion was particularly studied.

Electron microscopic histopathological studies done during the course of the present study revealed neuronal loss and gliosis in the epileptic focus. This is in accordance with the existing reports (Willmore and Rubin, 1982; Willmore et al., 1978). Neuronal loss and gliosis after FeCl₃ injection in the cortex are concurrent with the observed increase in lipid peroxidation products: 4-HNE and MDA (TBARS). To date we have not come across any report studying the ultrastructural changes in the iron epilepsy. At the ultrastructural level, in the neurons of epileptic animals, there was a loss of cytoplasmic homogeneity with the appearance of a number of vacuoles, increase in lysosomes and lipofuscin pigment, loss of endoplasmic reticulum, nucleolar budding etc. The attainment of irregular nucleolus contour due to the process of nucleolus budding in the neurons of cortex of epileptic animals could be the sign of an enhanced nucleolar synthetic activity under stressed conditions since the nucleolus is believed to be the main site for the synthesis of rRNA (La Velle and La Velle, 1975; Manocha and Sharma, 1977). The perineuronal accumulation of glial cells observed during the present study has also been reported to occur under stressful conditions (Olkowski and Manocha, 1972; Manocha and Sharma, 1978) such as age (Sharma et al., 1991). According to earlier reports iron-induced peroxidative reactions include formation of an area of gliosis, neuronal loss, dendritic alterations and neuronal ferrugination.
(Willmore and Rubin, 1982). Under stressful conditions, the association of glial cells with impaired neurons of epileptic cortex may be an adaptability of neurons to meet with the stressed environment developed in their cytoplasm due to the slowing down of protein synthesis. The slowing of protein synthesis during stressful conditions such as ageing has been known. The increased number of glial cells may be necessary for taking up the function of protein synthesis of the impaired neurons. The vacuolization observed, during the present study, in the cytoplasm of neurons of the epileptic focus may be due to the fact that stressed neurons have to meet the higher demands for protein and specific amino acids by consuming their cytoplasm (Kanungo et al., 1970; Zs-Nagy et al., 1979). The observed decrease in the extent of rough endoplasmic reticulum may relate to the fact that in the impaired neurones the protein synthesis is slowed down. The swelling of mitochondria observed during this study may be due to lipid peroxidation. Previous studies have reported this effect of lipid peroxidation on mitochondria (Kubo et al., 1984; Takei et al., 1994).

Besides electron microscopic observations, the present study also made some observations from magnetic resonance imaging studies. The magnetic resonance imaging observations showed the injection tract in untreated and the drug treated epileptic animals to be similar to that seen in the saline control animals. The magnetic resonance imaging pictures thus showed that the injection of iron chloride caused no extensive deleterious lesions in the brain.

Electrophysiological (electrocorticographic) recordings performed during the course of the present study revealed that intracortical injection of FeCl$_3$ in the rat brain parietal cortex caused the development of an epileptogenic focus in which spontaneous epileptiform electric activity persisted for several weeks. The epileptic electrophysiological activity began to appear within three days of the intracortical injection of FeCl$_3$. A conspicuous, intense recurrent epileptiform activity consisting of spikes and spike-wave complexes in the ipsilateral focus was observed by the days 8-15 of the intracortical injection of iron (FeCl$_3$). In the homotopic contralateral site also, an epileptic focus was found to develop in which the epileptiform activity was conspicuous although less intense as compared with that of the ipsilateral focus. The patterns and time course of the epileptiform activity were similar to those reported in many studies on the iron-induced focus in
rodents (Reid and Sypert, 1980; Colaisanti and Craig, 1973; Willmore et al., 1978 a and b; Willmore and Rubin, 1981; Singh and Pathak, 1990; Moriwaki et al., 1990 and 1992; Mizukawa et al., 1991; Shiota et al., 1989). Epileptiform discharges occurring in the electrocorticograms from the FeCl₃-induced focus compare well with the paroxysmal EEG epileptic activity observed in several types of genetically spontaneous epileptic rats (Vergnes et al., 1982; Sasa et al., 1988). The spike-wave complexes that occur in the iron-induced epilepsy also resemble the epileptiform activity observed by several research workers in other models of experimentally induced epilepsies (Colaisanti et al., 1974; Morocutti et al., 1986; Sugaya et al., 1988).

Besides electrocorticographic patterns accompanying the development of iron epileptogenic focus, the two other biochemical parameters which have been studied in the present work are, lipid peroxidation and the activity of the enzyme glutathione peroxidase. These two parameters are related to oxidative stress likely to be caused by iron. Alterations in the membrane lipid peroxidation and the glutathione peroxidase activity have been found to be related to epileptogenesis (Kurekci et al., 1995). The iron-induced epilepsy model continues to be of particular research interest since it models human post-traumatic epilepsy. There have been many studies on this model and investigations aimed at finding out the mechanisms underlying iron induction of epileptiform activity have not yielded conclusive information. In general the key mechanisms that mediate the conversion of normally functioning neurons into epileptiform activity-producing neurons are complex and are likely to vary with causative (etiologic) factors responsible for initiation of epileptogenesis. A number of neurochemical processes that may be related to mechanism of epileptogenesis have been reported to occur during iron induction of epilepsy. These include: membrane lipid peroxidation (Willmore et al., 1978 and 1983; Singh and Pathak, 1990); increased release of excitatory and other amino acids (Shiota et al., 1989; Hiramatsu et al., 1987; Janjua et al., 1990); decreased release of GABA (Zhang et al., 1989) which appeared to be a consequence of iron-induced lipid peroxidation; alterations in the cortical noradrenergic mechanisms - related alterations in cyclic-AMP (Hattori et al., 1990) etc.
The membrane lipid peroxidation resulting from iron-tissue interaction, however, is considered to be central to epileptogenesis in the iron model. Even some neurochemical changes that occur in iron epileptogenic focus such as a decrease in the release of GABA was found to be a consequence of lipid peroxidation (Zhang et al., 1989). There is enough relevant data concerning the possibility that membrane lipid peroxidation can alter the physiological status of membranes and can produce changes in the intrinsic properties of neuronal and synaptic membranes such that they would favour epileptogenesis. Lipids are involved in the biological functioning of membrane proteins including those involved in ion gating. Changes in the membrane lipid composition and lipid environment are known to lead to alterations in the allosteric properties of certain enzymes e.g. Na\(^+\), K\(^+\)-ATPase, acetylcholinesterase, Ca\(^{2+}\)-Mg\(^{2+}\) ATPases etc. (Housley et al., 1986).

Lipid peroxidation can alter membrane fluidity which in turn can influence opening and closing of excitable channels. In acetylcholine receptor functioning, it has been observed that changes in membrane fluidity can induce conformational changes associated with the opening and closing of receptor associated Na\(^+\) channels (Fong and McNamee, 1986). A study by Nelson and Delgado-Escueta (1986) revealed that the abnormalities of membrane fluidity are closely associated in the processes of epileptogenesis in freeze-lesion model in cat. An in vitro study concerning the electrophysiological correlates of lipid peroxidation damage in guinea pig hippocampus reports that both the synaptic efficacy and action potential generation are affected by damages resulting from lipid peroxidation (Pellmar, 1986). There is considerable evidence in the literature that membrane lipid peroxidation can significantly contribute to the development of paroxysmal electrophysiological membrane malfunction (Nelson and Delgado-Escueta, 1986). A study by Willmore et al., (1986) has evidenced the involvement of lipid peroxidation caused by iron salts in epileptogenesis in rat hippocampus.

In the present study, therefore, lipid peroxidation levels were studied in the whole homogenate of the ipsilateral as well as the contralateral epileptogenic foci. The data obtained during the course of the present study revealed that the increase in the lipid peroxidation (TBA-RS) in the ipsilateral epileptogenic focal tissue at different time points after FeCl\(_3\) injection, correlated with the development and
progression of the epileptic electrophysiological activity. Maximum levels of lipid peroxidation products were found on the day 8 after FeCl₃ injection. The levels persisted at significantly high levels through to day 28. Recurrent and well developed epileptic electrophysiological activity that had appeared by day 3 continued to occur thereafter.

The present data also showed some elevation of lipid peroxidation in the homotopic contralateral focus where epileptiform electrocorticographic activity occurred. However, these increases in lipid peroxides were much smaller than those in the ipsilateral focus. This is in accordance with the studies reported earlier (Singh and Pathak, 1990; Willmore et al., 1983).

The MDA content is often used to assess the extent of lipid peroxidation (Gutteridge and Halliwell, 1990) in a particular tissue, but more recent studies have suggested the involvement of other aldehyde products in the toxic effects of lipid peroxidation. 4-Hydroxynonenal (HNE) is thought to be one of the major toxic aldehyde by-products of lipid peroxidation (Benedette et al., 1984; Winkler et al., 1984). Its toxicity is attributed to its: inhibiting effect on enzyme activities (White and Reis, 1984), inhibition of DNA and protein synthesis (Poot et al., 1988), and lowering of cellular glutathione levels (Haenan, 1987). Reactive aldehydes (Boehme et al., 1977) released by peroxidation (Pryor and Stanley, 1975) form adducts with thiols and amino acids or their esters (Bidlack, 1973) and these insoluble compounds are known to form conjugated fluorescent Schiff’s bases (Chio and Tappe!, 1969; Fletcher et al., 1973). Considering the cellular toxicity that 4-HNE and MDA can cause, it was suggested that estimation of 4-HNE along with MDA can give a better understanding of the lipid peroxidation undergone by the tissues (Halliwell and Gutteridge, 1987). Thus in the present study, to have a more comprehensive assessment of the degree of lipid peroxidation, the levels of 4-HNE were also measured in the ipsilateral and contralateral epileptogenic foci.

In the present study, 4-HNE levels were found to significantly increase at day 8 after FeCl₃ injection and gradually further increased thereafter. Similar significant increase in 4-HNE levels also occurred in the contralateral focus. The quantity of fluorescent products of lipid peroxidation was measured in acute epileptogenesis induced by iron in a study by Willmore et al.(1986). They also observed a similar accumulation of fluorescent lipid peroxidation products. However, their study was done on animals.
decapitated only two hours after hippocampal injection of iron salts; and in another experiment they found the levels of 4-HNE to increase in the epileptic amygdala (Triggs and Willmore, 1994). Data from the present study, however, provide more extensive information on the occurrence of lipid peroxidation in the iron focus as the levels of lipid peroxidation products in the epileptogenic focus were assessed through a period of 28 days. The increase in levels of 4-HNE observed during this study was concurrent with the observed increase in MDA levels and the build up of the focal epileptic electrographic activity as well.

In the present study the activity of glutathione peroxidase (GPx), an important antiperoxidant enzyme, was also measured in the epileptogenic focus. The data obtained showed that the activity of GPx in the epileptogenic focus was only slightly elevated. This low elevation of the enzyme activity observed in the present study is in agreement with previous reports (Singh and Pathak, 1990). The antiperoxidant enzyme GPx oxidizes reduced glutathione, thus making available a detoxification potential for the neutralization of peroxides (such as \( \text{H}_2\text{O}_2 \), lipid peroxides). Several studies have shown that GPx is of major importance in the detoxification of peroxides in the brain (Hothersall et al., 1982; Kish et al., 1985). GPx can thus offer significant protection against peroxidative damage. The low elevation of GPx activity found in the present study was concurrent with the high increase in lipid peroxidation during the iron epilepsy. Thus it can be argued that the high levels of lipid peroxides are accumulated in the epileptic focus because somehow GPx fails to be stimulated sufficiently. The marked deficiency of GPx activity in the epileptogenic focus may, therefore, be an important factor in the pathogenesis of iron-induced experimental epilepsy. In other studies also it has been shown that decreases in the activity of glutathione peroxidase are associated with elevation of lipid peroxides. For example Yalcin et al. (1986) found stimulation of lipid peroxidation together with an impairment of glutathione - dependent defence system in the liver of rats repeatedly treated with carbon tetrachloride (CCl\(_4\)). Their results showed a significant decrease in GSH levels and GPx activity, thus indicating that the increased hepatic lipid peroxidation is at least in part due to the absence of the protective action of GPx.

The present study was designed to determine whether various antiperoxidant drugs have antiepileptic effects against iron-induced epilepsy in the rat. The drugs studied were: \( \alpha \)-tocopherol, verapamil, flunarizine, ethosuximide and sodium
valproate. Their effects were assessed by changes produced in the epileptic electrographic activity (ECoG), lipid peroxidation and the glutathione peroxidase activity after drug administration.

5.2. α-Tocopherol

Treatment of epileptic animals with α-tocopherol caused complete suppression of the epileptogenic electrocorticographic activity in the epileptogenic focus together with a significant decline in the levels of lipid peroxides in the ipsilateral and contralateral epileptogenic foci. The drug treatment also increased the activity of glutathione peroxidase in the epileptogenic focus. The protective effect of the antioxidant α-tocopherol was observed at ultrastructural level also. The treatment with α-tocopherol resulted in reappearance of the extensive rough endoplasmic reticulum and myelination. Furthermore there was a decrease in the number of lysosomes, and an improvement in the cytoplasmic and nuclear homogeneity and vacuolation etc. Since it is known that lipid peroxidation can produce cytological damage, the cytological effects of α-tocopherol as observed in the present study can be attributed to the drug's anti-lipidperoxidative effects (Kanungo et al., 1970). In previous studies only the pretreatment of rats or cats with α-tocopherol was studied and was shown to prevent histological perturbations, necrosis and gliosis caused by iron salts injection into the rat isocortex (Willmore and Rubin, 1981) or cat spinal cord (Means et al., 1983).

In a study by Levy et al. (1990), vitamin E pretreatment was found not to completely prevent the iron-seizures but it only delayed their onset. In other studies, however, vitamin E pretreatment was reported to prevent iron seizures only in a large number of experimental subjects (Willmore et al., 1978; Rubin and Willmore, 1980) but not in all animals. The data presented in the present study thus significantly extends these findings regarding the α-tocopherol effects on iron epileptogenesis since our data show that the drug can produce its antiepileptic effects even against an active epileptogenic focus. The results of the present work have clearly shown that prolonged treatment with vitamin E completely suppresses the iron-induced seizures. The failure noted in the previous studies was probably
due to the use of short term doses. Our data are of relevance to some clinical studies where it was found that the vitamin E when used as adjunct or add-on therapy was effective in treatment of epilepsy. The finding from the present work that α-tocopherol is also effective against the epileptic activity of the contralateral epileptic focus is also of interest. From this, it would appear that the vitamin E on its own could be effective against spontaneous seizures (Levy et al., 1990). Our data also indicate that the anti-lipidperoxidative effects of α-tocopherol will partly be due to the drug's stimulation of the enzyme glutathione peroxidase. The deficiency of glutathione peroxidase has been found to be of causal significance in clinical epilepsies (Kurekci et al., 1995). The efficacy of vitamin E therapy found against clinical seizures, therefore, may also be attributed to the drug's effect on glutathione peroxidase activity.

In summary, the data derived from the present study show that post treatment of iron-induced epileptic animals with α-tocopherol is efficacious in: suppressing epileptic electrocorticographic activity; decreasing lipid peroxidation; and elevating glutathione peroxidase activity in the epileptogenic focus. The data also revealed that these antiepileptic effects were also manifest against contralateral focus epileptiform activity indicating thereby that at least to some extent α-tocopherol (vitamin E) has antiepileptic effects also against epileptiform activity arising due to causes other than metal-tissue interaction.

Although deficiency in antioxidant enzymes such as the enzyme glutathione peroxidase appears to be causally implicated in some forms of clinical epilepsies and such cases are treated with usual AEDs, knowledge about the effect of antiepileptic drugs on antioxidant enzymes is poor (Kurekci et al., 1995). In view of this, the results of the present work on the effect of antiepileptic drugs on glutathione peroxidase in iron-induced epileptogenic tissue are of interest.

5.3. Verapamil

Verapamil is a known anticonvulsant drug having Ca$^{2+}$ channel blocking properties. In contrast to some Ca$^{2+}$ antagonists such as nimodipine and nifedipine which act on the extracellular side of the Ca$^{2+}$ channel, verapamil acts on the intracellular side of the channel (Kamal et al., 1990). It is a rather weak
anticonvulsant which may be due to its poor blood-brain crossing properties. That is why sometimes its antiepileptic efficacy is considered doubtful. In vitro studies have shown demonstrably effective antiepileptic effects of this drug (Speckmann et al., 1990). Similarly, intracerebroventricularly administered drugs have been found more efficacious. In some studies where intraperitoneal administration of verapamil was adopted, weak anti-epileptic effects were found (Kamal et al., 1990). The results of the present work, however, clearly showed that verapamil when given intraperitoneally suppressed the epileptiform electrocorticographic activity, decreased the lipid peroxidation and elevated the glutathione peroxidase activity in the ipsilateral iron-induced epileptogenic focus. In the contralateral focus also the drug showed similar effects. The anti-lipidperoxidative effects of verapamil in the iron epileptic focus appear similar to its effects in other tissues. For example, Robak and Dunnier (1986) found verapamil inhibition of hepatic membrane peroxidation in a dose dependent fashion. The drug binds to membranes and thus provides protection against lipid peroxidation. Arouma et al. (1991) also reported that verapamil inhibited peroxidation of the rat liver microsomes induced by FeCl₃ and ascorbic acid. They too observed, that antioxidant effects of the drugs tested, are likely to be exerted only against lipid peroxidation and that also only if the drug accumulates in membranes to a sufficiently high concentration. Reddy et al. (1996) and Sugawara et al. (1996) too have shown that verapamil inhibits iron dependent lipid peroxidation by its antioxidant properties, apart from blocking calcium influx. Verapamil is also known to prevent calcium dependent protein kinase activation and thus leading to a limiting of the conversion of xanthine dehydrogenase (XD) to xanthine oxidase (XO) and thus reducing lipid peroxidation (Vanella et al., 1992).

In summary the present data have demonstrated significant antiepileptic effects of verapamil in iron focus and these effects are mediated by reduction of lipid peroxidation and the elevation of glutathione peroxidase activity. The present data further confirm that intraperitoneally administered verapamil does show antiepileptic effects.
5.4. **Flunarizine**

Flunarizine is a potent calcium channel antagonist with antiperoxidant properties. The drug differs from a number of antiepileptic drugs in its mechanism of action (Bebin and Bleck, 1994). The present results showed that on treatment with flunarizine, levels of TBA dropped to almost control levels. The contralateral epileptogenic focus too showed a decrease in lipid peroxidation. Levels of 4-HNE were also significantly reduced by flunarizine. Flunarizine enhanced the activity of glutathione peroxidase both in the ipsilateral and the contralateral foci. The enzyme activity increased from the day 8 after iron injection, and then progressively increased thereafter. This was the case in the contralateral focus also. The drug also suppressed the electrographic seizure activity with a marked change. The frequency and amplitude of the electrocorticogram decreased both in the contralateral and the ipsilateral foci and this change was apparent from the first day of treatment itself.

The influx of calcium into neurons plays an important role in triggering of epileptic activity. Flunarizine has been believed to act as an antiepileptic by inhibiting influx of calcium ions (Speckmann and Walden, 1986; Cousin et al., 1993), (by blocking the voltage dependent calcium channels) and diminishing neuronal hyperexcitability. Flunarizine appear to actually blocks T-type Ca\(^{2+}\) channels but it does not affect the normal function of voltage -dependent Ca\(^{2+}\) channels and therefore synaptic transmission remain unaffected. Thus, in similarity with organic Ca\(^{2+}\) antagonists it does not depress non-epileptic neuronal activity (Speckmann et al., 1990). Flunarizine also has an inhibitory effect on glutamate release (Cousin et al., 1993). Part of its anticonvulsant activity may be due to its effects on sodium channels (Wauquier et al., 1986). Flunarizine is also known to be a free radical scavenger. Flunarizine can reduce free fatty acid release and like tocopherol is a more potent antioxidant in the mitochondrial suspension than in the homogenate suggesting that penetration of this drug into a precise site may be a critical feature of its inhibition of lipid peroxidation (Kubo et al., 1984). Flunarizine is known to enhance the anticonvulsant effect of other antiepileptic drugs such as carbamazepine, diphenylhydantoin in animal seizure models (Joseph et al., 1998). The drug has been shown to suppress epileptiform
electrophysiological activity in a variety of seizure models (Schulze-Bonhage et al., 1994).

In the present study, on the whole, flunarizine appears to show a higher potency to reduce epileptogenesis. The present results demonstrate that flunarizine has strong antiepileptic effects in iron-induced epileptogenic focus, and the drug's antiepileptic effects may be due to its anti-lipidperoxidative and calcium channel blocking properties. The data obtained in the present experiments are thus in accord with the drug's known general anti-lipidperoxidant and Ca\(^{2+}\) - channel antagonist properties.

5.5. **Ethosuximide**

The drug ethosuximide is known to be effective against absence seizures, and has been tested in a number of animal models (Stringer, 1994). The present study demonstrates that the drug is effective against iron-seizure model. The data derived from the present experiments showed that the drug decreased the lipid peroxidation levels (measured by TBA-RS) in the ipsilateral focus. The levels came down to control levels. Decreases were seen after day 1 of the drug administration. The levels were then maintained at a low level indicating that lipid peroxidation was lowered. In the contralateral focus too a similar decrease in lipid peroxide levels was conspicuous. The levels of 4-HNE also declined in both the ipsilateral and contralateral foci. The decrease in levels occurred progressively. As the lipid peroxide levels fell the levels of GPx were found appreciably elevated as compared to those of iron-induced epileptic animals. The increase in GPx levels will be consistent with the massive lowering of lipid peroxidation levels. It would thus appear clear that the antiepileptic action of the drug in this model is associated with alterations in lipid peroxidation mechanisms. Ethosuximide also greatly suppressed the electrographic seizure activity. Ethosuximide is known to have an efficacy in suppressing spike and wave discharges, and also T-type thalamic calcium currents, as seen in anti-absence epilepsy. The drug has also been shown to prevent the spread of seizure activity through cortical pathways (Fromm et al., 1980); it has desynchronizing effects on EEG activity (Wenzel et al., 1971), and it has also been found to increase slow wave activity in cortical EEG together with a
behavioural sedative effect (Mirski and Ferrendelli, 1986). The mechanism of action of the drug is presumed to be related to its action on Ca$^{2+}$ currents particularly thalamic currents. The drug's effects on lipid peroxidation and antioxidative enzymes have not been studied so far. The present data demonstrate that ethosuximide also has anti-lipidperoxidative properties. In this respect the drug would appear to be similar to other Ca$^{2+}$ channel blocking agents. The present data thus provide additional new information about the mechanism of action of ethosuximide that the drug is an anti-lipidperoxidant.

5.6. Valproate

Valproate is a known antiepileptic drug. Clinically it is effective against both generalized and convulsive seizures (Stringer, 1994). It has a broad spectrum of anticonvulsant activity in animal seizure models also. It has multiple mechanisms of action in central neurons. Besides its action on GABA system, it blocks voltage-dependent Na$^+$ channels. In addition, recent work has shown that it is capable of reducing the low threshold Ca$^{2+}$ currents known as T currents (Kelly et al., 1990). These currents may also be implicated in its antiepileptic effects. Valproate on entering the brain predominantly distributes in the extracellular space. It acts on the extracellular side of the neuronal membrane and enters the intracellular compartment also and that is why its effects persist (Loscher and Honack, 1995). The results of the present study show that valproate reduced the epileptiform activity in ECoG and it acts as an anti-peroxidant as the drug lowered the levels of TBA-RS markedly in the ipsilateral and in the contralateral foci. 4-HNE, the other parameter studied for assessing levels of lipid peroxidation, was also found to be decreased by the drug. The drug also induced an increase in the levels of GPx in both ipsilateral and contralateral foci. The present data thus show that valproate exerts anti-lipidperoxidative action in the epileptic tissue.

Valproate has been indicated to influence lipid peroxidation, though it itself is not an antioxidant (Nikushkin et al., 1993). It has been found to induce the activity of GPx in the plasma of human epileptic patients (Kurekci et al., 1995) probably by induction of hepatic synthesis of glutathione peroxidase. The data obtained in the present study in addition demonstrate that valproate has anti-
lipid peroxidation action in the epileptogenic neural tissue. The data derived from the present experiments would be in accord with the role of glutathione peroxidase in some forms of clinical epilepsies and the anti-lipid peroxidative effects of AEDs. The experimental data from the present experiments thus support results of clinical studies showing a causal role for glutathione peroxidase in some forms of epilepsies.

5.7. Ultrastructural effects of the antiepileptic drugs in iron-induced focus

The electron microscopic data obtained in the present study demonstrated that the antiperoxidant antiepileptic drugs reversed the deleterious structural changes in the lesion induced by iron. The most prominent change seen in the drug-treated animals was the reappearance of rough endoplasmic reticulum (RER) and Golgi bodies in large numbers. Free ribosomes also became conspicuous. This may be because protein synthesis reoccurs at a higher rate and these cells somewhat normalize their activities. The cytoplasm regained its homogeneity and the vacuolization decreased. The lysosomes showed a reduction in their number signifying that the hydrolytic activity greatly declined. The mitochondria also showed a return to normal size except in the ethosuximide treated animals that still showed swollen mitochondria. The chromatin became dispersed and the dense chromatin apparent in epileptic animals was no longer visible. The nucleolus too showed some activity, the dark spots visible being indicative of that.

In conclusion, the data derived from the present experiments demonstrated that iron-induced epileptiform activity develops because of increases in TBARS and 4-HNE and an insufficiency in the glutathione peroxidase activity in the epileptogenic focus. It is of interest to note that anticonvulsants which did not have antiperoxidant actions were reported to be not efficacious in preventing trauma-induced seizures and the need for more specific formulation of pharmacologic agents to prevent or treat post-traumatic epilepsy was indicated (Willmore and Rubin, 1981). Therefore, the results of the present study have provided significant information regarding the possibility of the use of anti-peroxidant drugs in the treatment of post-traumatic epilepsy.