DISCUSSION
SUMMARY OF THE RESULTS:

The results of this study suggested the following:

1] REM sleep deprivation caused an increase in the rat brain AChE activity.

2] The increase in AChE was expressed first in the medulla.

3] REM sleep deprivation affected both the forms of AChE in the rat brain. The change was seen primarily in the brain stem regions. There was an increase in the activity of the membrane bound form and a decrease in the activity of free or the soluble form of AChE.

4] REM sleep deprivation caused a decrease in the rat brain MAO, MAO-A and MAO-B activities.

5] The effect on MAO-A activity was much stronger than that on the MAO-B activity. Pons was the first area/site to be affected.

6] Long term REM sleep deprivation caused an increase in the activity of hexokinase in the rat brain. The change was seen primarily in the brain stem and cerebellum.

7] Short term REM sleep deprivation did not have any effect on the activity of hexokinase in any of the brain stem regions.

8] REM sleep deprivation caused a decrease in the activity of glucose-6-phosphatase in the rat brain.

9] 24 h REM sleep deprivation affected the activity of glucose-6-phosphatase maximally, in the cerebrum whereas four days deprivation showed an increase as compared to one day deprivation.

8] REM sleep deprivation affected the activity of the enzyme 5’-Nucleotidase in the cerebrum of the rat brain. No other area showed any significant alterations.

9] Long and short term REM sleep deprivation caused an increase in the membrane fluidity in the cerebrum and the brain stem.

10] REM sleep deprivation had no effect on the levels of total phospholipids in any region of the brain nor was any change observed.
in the levels of individual phospholipids after REM sleep deprivation. The activities of the enzymes lactate dehydrogenase and glucose-6-phosphate dehydrogenase, in the rat brain were also unaffected after long term REM sleep deprivation.
The results obtained in this study are discussed in detail under the following under the following sub headings:

A] The validity of the methodology used for REM sleep deprivation
B] Effect of REM sleep deprivation on the activities of AChE
C] Effect of REM sleep deprivation on the activities of different forms of AChE
D] Effect of REM sleep deprivation on the activities of MAO, MAO-A and MAO-B
E] Effect of REM sleep deprivation on the activities of the enzymes involved in the metabolism of glucose
F] Effect of REM sleep deprivation on the activity of 5'-Nucleotidase
G] Effect of REM sleep deprivation on membrane fluidity.
H] Effect of REM sleep deprivation on phospholipids.

A] THE VALIDITY OF THE METHODOLOGY USED FOR REM SLEEP DEPRIVATION:

The results obtained in this study were the specific effects of REM sleep deprivation and unlikely to be artifact induced by the method used for deprivation because the flower pot method, preferred in this study, is probably the ideal method and the most widely used for REM sleep deprivation (Bowers et al. 1966, Mendelson et al. 1974, Vogel, 1975, Van Luijtelaar and Coenen, 1976, Hicks et al. 1977, Yanick and Radulovacki, 1987, Oniani et al. 1988, Coll-Andreu et al. 1989, Mallick et al. 1989, Mallick and Thakkar, 1991, Thakkar and Mallick 1991). Furthermore, to rule out the possibilities of other non-specific effects, in addition to the FMC and R experiments, LPC, SW and RM experiments were carried out. The LPC rats served as control for environmental non-specific factors while the RM rats served as control for the immobilization stress, if
any, involved due to restriction of movement to the E animals. The movement was restricted as long as a fortnight. The size of the platforms were chosen corresponding to the weight of the rats based on earlier reports (Mendelson et al. 1974, Yanick and Radulovacki, 1987). The effect was not due to increased muscular activity on smaller platforms because the SW control rats did not show any change. Two sets of swimming experiments were done with an idea that since it is difficult to equate the stress, if at all, induced by deprivation and due to swimming, if the effect on the enzyme activities would have been due to stress, the two activities would be have been different.

B] EFFECT OF REM SLEEP DEPRIVATION ON THE ACTIVITY OF AChE:

The results of this study suggested that REM sleep deprivation caused an increase in the rat brain AChE activity. Eight days REM sleep depletion increased AChE activity significantly in the whole brain homogenate as well as in the brain stem and the cerebrum. However, four days deprivation induced a significant increase in AChE activity in brain stem only. The activity in the cerebellum was never affected. Though, AChE activity was not significantly affected after 48 hrs of REM sleep deprivation in the brain stem as a whole, however, it was affected in the medulla. Even after 24 and 48 hrs deprivation caused a significant increase in the activity of the medullary AChE only. Pons and midbrain were never affected.

Results of this study get the supporting evidence from the fact that the levels of ACh are increased during REM sleep (Jasper and Tessier, 1971, Gadia-Circia et al. 1973, Hanranth and Venkatakrishna-Bhatt, 1973), and there is a decrease in the levels ACh after REM sleep deprivation (Bowers et al. 1966). Alteration in the AChE activity with changes in the levels of ACh has also been reported.
(Rosenberry, 1975). Hence, REM sleep deprivation induced significant increase in AChE activity in the rat brain, may be explained as follows: First, the deprivation might have a direct effect to increase the activity/level of AChE in the brain. Though the mechanism of increase is yet to be investigated, it may be supported by earlier report that cholinesterase inhibitor, which is likely to reduce the level of AChE, have shown to increase the REM sleep (Amatruda et al. 1975, Jouvet, 1975, Hobson et al. 1983, Gillin et al. 1985). Second, due to REM sleep deprivation there might have been a tendency for an increase in the level of ACh (possibly due to REM pressure) which led to an increase in AChE activity and this may cause the decrease in ACh in REM deprived rat brain (Bowers et al. 1966) Third, increase in the heart rate and energy expenditure (Bergmann et al. 1989, Kushida et al. 1989) on REM sleep deprivation may be due to reduced level of ACh as the result of an increase in the activity of AChE. Thus REM sleep deprivation may be a withdrawal of para- sympathetic effect. Fourth, the present finding may explain earlier observations that REM sleep is disturbed in pathological states where ACh or AChE levels are affected (Casal do, 1969, Papazian, 1976). Fifth, this finding supports the cholinergic mechanism in REM sleep (Jouvet, 1975, Gillin et al. 1985). It is interesting that the increase in the AChE activity in the whole brain could be seen after 8 days and not 4 days deprivation; in the brain stem after 4 days and not 2 days deprivation; and in the medulla after 1 & 2 days deprivation. It is tempting to hypothesize that REM sleep deprivation induced increase in AChE starts at the medulla and spreads to other regions of the brain. Though it needs to be confirmed, the activity did not change in the cerebellum probably goes against the hypothesis.

The increase in the activity of medullary AChE may be supported
by earlier reports that REM related atonia and EEG desynchronization are mediated through the medullary cholinergic mechanism (Sakai, 1984, Schenkel and Siegel, 1989). Hence, the results obtained in this study may represent a step forward in settling the issue regarding the precise area of the brain stem responsible for generation of REM sleep signs through the cholinergic mechanism (Amatruda et al. 1975, Jouvet, 1975, Hobson et al. 1983, Sakai, 1985, Gillin et al. 1985, Gnadt and Pergram, 1986, Shiromani et al. 1987, Jones, 1991).

C) EFFECT OF REM SLEEP DEPRIVATION ON THE MOLECULAR FORMS OF AChE:

The results of this study suggest that REM sleep deprivation affects both the forms of AChE in the rat brain. The change was seen primarily in the brain stem regions. There was an increase in the activity of the membrane bound form and a decrease in the activity of free or the soluble form of AChE. Those forms of AChE did not show any significant alteration in the cerebrum after REM sleep deprivation.

Membrane bound form of AChE has been proposed to be present in the nerve endings and primarily responsible for breakdown of ACh (Sirvio et al. 1989). Increase in the activity of the membrane bound form of the enzyme fits well with the earlier studies where it has been shown that the ACh decreased (Bowers et al. 1966) on REM sleep deprivation. It supports the involvement of the cholinergic mechanism in the generation and regulation of REM sleep (Jouvet, 1975). Membrane bound AChE appears to be more vulnerable to REM sleep deprivation as also observed during post-natal development as well as during ageing (Sung and Ruff, 1983, Muller et al. 1985, Sirvio et al. 1989). During ageing there is a reduction in REM sleep, while in infancy there is a higher percentage
of REM sleep (Jouvet-Monnier et al. 1969) where the activity of the membrane bound form is reported to decrease and increase, respectively. However, in this study it has been observed that REM sleep deprivation induced an increase in the activity of the membrane bound form of AChE. This may be attributed to the possibility that REM sleep deprivation might induce a REM pressure within the system and thereby causing an increase in the enzyme activity. The increase in the activity of the membrane bound form and a decrease in the activity of the free or the soluble form may reflect its conversion to the bound form (Fig. 5) or it may not be related to change in the ACh.

Pons is the primary site to be affected by REM sleep deprivation. This may be due to the fact that pontine brain stem region is involved in the generation and regulation of REM sleep (Hobson et al. 1986, Jones, 1991). Short term (48 h) REM sleep deprivation was enough to cause an increase in the activity of the membrane bound form of AChE in the pons and a decrease in the free form in the medulla though it did not affect either of the forms of AChE in the midbrain. Thus, the changes in the enzyme activities in the pons and the medulla support the ponto-medullary interaction mechanism for the generation of REM sleep (Hobson et al. 1986, Jones, 1991).

It is interesting to note that there is an increase in the activity of the membrane bound form and a decrease in the activity of the free form but the opposite was never observed. Though the mechanism of REM sleep deprivation-induced alterations in the enzyme activity is yet to be studied, the following possibilities may be postulated: 1) as the membrane bound form (10S) is located mainly in the nerve terminals and the free form is cytosolic, it is possible that REM sleep deprivation might cause a conversion of the free form of the enzyme
to the bound form (possibly due to REM pressure) which might lead to REM sleep deprivation induced reduction in the level of ACh. As reflected in Fig. 6 there is a possibility of the conversion since the bound to free ratio is always positive. However, as evidenced from Fig. 1 the conversion, if at all, is unlikely to be proportional and the increase in the activity of the bound form is more than the decrease in the activity of the free form; 2) the deprivation might induce an alteration in the membrane per se which may cause an increase in the activity of the membrane bound AChE. It may be indirectly supported with the observation that other membrane bound enzyme viz. Na-K ATPase (Gulyani and Mallick, 1993) is also affected after REM sleep deprivation; 3) It may also be possible that REM sleep deprivation induced changes in the level of hormones or small molecular weight proteins (Inoue, 1989) may affect the enzyme activity directly or indirectly through changes in the membrane. At this stage it is difficult to comment on the kinetics of the different forms of AChE which may also be affected on REM sleep deprivation.

D] EFFECT OF REM SLEEP DEPRIVATION ON THE ACTIVITIES OF MAO, MAO-A AND MAO-B:

The results of this study showed that four days REM sleep deprivation though caused a decrease in the rat brain MAO and MAO-A activities, MAO-B did not show any significant change. However, the latter was affected after short term REM sleep deprivation. In the majority of instances the effect on MAO-A activity was much stronger throughout. The decrease in the enzyme activity was proportional to the number of days of deprivation - longer was the deprivation, lesser was the enzyme activity. However, after short term (1&2 days) deprivation the enzyme activity though showed either a decrease or no change in the pons and the midbrain, showed
an increase or no change in the medulla. The increase was reflected in MAO and MAO-A activities in the medulla only while all other areas, where ever there was a change, showed a decrease. MAO-B activity when ever was affected, showed a decrease. Though medulla was the first site to be affected where the enzyme activity increased even after 24 hr REM sleep deprivation, pons was the first area/site where the enzyme activity showed a decrease but only after 2 days deprivation. The enzyme activities did not change in the control rats and the deprivation induced alteration in the enzyme activities returned to the baseline after recovery.

The significant decrease in MAO activity in rat brain on REM sleep deprivation as observed in this study may be explained and supported by earlier studies. The deprivation might have a direct effect to decrease the MAO activity in the rat brain or the effect may be mediated through the changes in the release or levels of NE (Houslay and Tipton, 1973, Turner et al. 1974), which is reported to be affected on REM sleep deprivation. Though it is tempting to put forward that MAO activity may alter parallel with NE release, there are reports suggesting that the activity of MAO is unlikely to be related to the release of NE (Tipton, 1979, Finberg and Youdim, 1983). The deprivation might cause an increase in endogenous MAO inhibitor viz. tribulin (or alike) which is known to increase at least on stress (Armando et al. 1989, Bhattacharya et al. 1989). Though the mechanism of decrease is yet to be investigated, it may be supported by earlier report that MAO inhibitors and monoamine uptake blockers, which are likely to increase the level of NE, are known to decrease REM sleep (Vogel et al. 1990). It has been found in this study that the MAO activity decreased in those brain areas where either NE-ergic neurons or their projections are relatively more. The NE-ergic neurons are located in ponto-mesencephalic region with a
maximum concentration in the rat locus coeruleus (Jacobs, 1986) and the cerebrum/ cortex receives a bulk of its projections (Dahlstrom and Fuxe, 1964, Ungerstedt, 1971). The pontine NE- ergic neurons (REM-OFF) are known to be affected by REM sleep deprivation (Mallick et al. 1989). Therefore, probably the MAO activity in the cerebrum and the pons get affected maximally. However, on short term REM sleep deprivation medulla was the first area for MAO activity to be affected and that was an increase i.e. opposite to that of long term deprivation. Since brain stem also receives NE inputs from areas other than the locus coeruleus (Fristchy and Grzanna, 1990), the possibility of NE-ergic fibers projecting from areas in the brain stem other than locus coeruleus in mediating such changes cannot be ruled out.

The alterations in MAO-A and MAO-B activities on REM sleep deprivation are interesting. The stronger influence (a decrease) on the former supports the involvement of norepinephrinergic mechanism in REM sleep (Jouvet, 1972, Hobson et al. 1986, Siegel, 1989, Jones, 1991). It is difficult at present to comment if the deprivation induced alteration in the MAO-A activity is a primary effect, which ultimately affects the level of NE or secondary to changes in the turnover of NE (Stern et al. 1971). The MAO-A activity is known to be affected by small peptides and hormonal levels (Tipton, 1979, Youdim et al. 1989, Tomaszewicz et al. 1991). Since REM sleep deprivation is known to affect hormonal levels (Kushida et al. 1989) and is likely to affect the levels of sleep substances (Inoue, 1989, Obal et al. 1989), the changes in the MAO-A activity may be a secondary effect.

The decrease in MAO-B activity reflects that the REM sleep deprivation probably affects the biogenic amines in general (Tipton, 1979, Finberg and Youdim, 1983) in the system. It may be supported
from earlier reports that the MAO-B activity increases in Alzheimer’s disease (Oreland and Gottfries, 1986) where the REM increases (Bliwise et al. 1990). It has been found that the MAO-A and MAO-B are affected differentially. Short term REM sleep deprivation influenced either the MAO-A or the MAO-B activity within the different regions of the brain stem (midbrain, pons and medulla). This may possibly be due to local interaction among different neurotransmitters and small peptides viz. Somatostatin, VIP, Substance P, etc. (Jones, 1991) which have been demonstrated to coexist in the brain stem neurons (Holets, 1990) and influence REM sleep (Inoue, 1989, Obal et al. 1989).

It must be noted that after short term (1 & 2 days) REM sleep deprivation the first area to be affected is the medulla where the MAO-A activity showed a tremendous increase which was reduced and reversed on longer deprivation. Similar short term deprivation (24 hr) has been reported to increase the AChE activity in the medulla only (Mallick and Thakkar, 1991). On putting all these facts together it may be suggested that the integration of the aminergic and cholinergic mechanisms for the generation of REM sleep (Hobson et al. 1986) may be taking place some where in the medullary region of the brain stem and short interneurons containing low molecular weight peptides may possibly be involved in the modulation of such a process. The medullary REM-ON neurons (Kanamori et al. 1980, Sakai, 1985a) might be involved in mediating and/or initiating such response. Normally cessation of firing of the NE-ergic REM-OFF neurons during REM sleep probably upregulates and/or resensitizes the NE receptors (Siegel and Rogawaski, 1988). Reduction of firing of those REM-OFF neurons probably is a step towards compensatory mechanism (Mallick et al. 1989). Since the REM-OFF neurons do not stop firing on REM sleep deprivation, it is likely to cause a
continuous release of NE. Increase in MAO-A activity in the medulla probably tries to reduce the NE level as compensation towards normal situation. However, when that fails due to prolong deprivation the activities of MAO-A as well as MAO-B decrease throughout the brain though the former is affected more. Alternatively, the changes seen in the MAO activity, as observed in this study, may probably be a reflection of a different time course of NE release by the REM-OFF NE-ergic neurons in relation to REM sleep deprivation.

The findings of this study support the involvement of aminergic mechanism in REM sleep regulation (Hobson et al. 1986).

EFFECT OF REM SLEEP DEPRIVATION ON THE ACTIVITIES OF ENZYMES INVOLVED IN GLUCOSE METABOLISM:

The results of this study showed that REM sleep deprivation first significantly affected the activity of G-6-Pase while the activity of hexokinase was affected latter, though the activities of LDH and G-6-PDH were not affected significantly. The two enzyme (hexokinase and G-6-Pase) activities were affected in opposite directions and the maximum effect was seen in the brain stem. Long term (four days) REM sleep deprivation caused a significant increase in the activity of hexokinase in all the areas of the brain. Glucose is the primary source of energy for the brain and hexokinase is the first rate limiting enzyme in the glycolytic pathway. REM sleep deprivation induced alteration in the activity of hexokinase is likely to enhance metabolism of glucose and thereby an increase in energy production. REM sleep is reduced in diabetes (Kapas et al. 1991) where the glucose metabolism is decreased. REM sleep deprivation induced increase in hexokinase activity probably indicates that a REM pressure may be induced in the system which in turn sets the mechanism (i.e. increase in hexokinase activity) for increased glucose
metabolism. It may be supported by the fact that glucose metabolism in the brain increases during REM sleep. Increased activity of hexokinase in the brain stem may be possibly because it is the region involved in the regulation and generation of REM sleep (Sakai, 1985, Siegel, 1989). The majority of the brain areas which show the changes in the glucose metabolism during REM sleep, are proposed to be involved in the regulation of REM sleep (Lydic et al. 1991). On the other hand, short term (48 h) deprivation had no effect on the activity of hexokinase.

Short and long term REM sleep deprivation caused a significant decrease in the activity of the enzyme G-6-Pase. Brain stem was the region which showed the maximal decrease in G-6-Pase activity, while the same showed a maximum increase in the activity of hexokinase. This enzyme is involved in the conversion of glucose-6-phosphate to glucose. An increase in its activity during sleep, when glucose metabolism is at its lowest, has been reported (Anchors and Karnovsky, 1975). It has been proposed that there is an increase in the cell to cell transfer of glucose during sleep (Karnovsky, 1991) probably leading to a decrease in the metabolism of glucose. The decrease in the activity of G-6-Pase is likely to reduce the glucose transfer and thus, may possibly be responsible for REM sleep deprivation induced increase in the energy expenditure. The decrease was more after short term REM sleep deprivation and was reduced (less decrease) on long term deprivation, an opposite effect as compared to that of an increase in hexokinase activity (mentioned earlier). REM sleep deprivation initiates an increase in the energy expenditure. Initially after short term deprivation the increase was caused by a decrease in the reshuffling of glucose, due to a decrease in G-6-Pase activity, while with increase in deprivation there was a compensatory increase in the aerobic glycolytic flux due to an
increase in the activity of hexokinase. This may be supported by previous reports that REM sleep deprivation causes a decrease in the levels of glycogen in the brain (Karadzic and Mrsulja, 1969) and on long term REM sleep deprivation there was an increase in the degradation of fats and proteins (Kushida et al. 1989). It may also be possible that deprivation induced REM pressure may cause a decrease in the activity of G-6-Pase.

The mechanism for the alterations in the enzyme activities, after REM sleep deprivation is difficult to establish from this study. Nevertheless, the following may be put forward as the possible mechanisms. First, REM sleep deprivation is likely to cause an increase in the REM pressure which in turn may be involved in causing the alterations in the enzyme activities. This may be supported by the fact that glucose metabolism increases during REM sleep (Ramm and Frost, 1986, Sawaya and Ingvar, 1989, Lydic et al. 1991) and infusion of glucose, pyruvate and lactate are reported to induce REM sleep (Kawakami and Yoshida, 1965). Second, it may be possible that REM sleep deprivation is known to induce changes in the level of hormones and neuropeptides (Inoue, 1989) which in turn might affect the enzyme activities directly or indirectly. Third, the deprivation induced increase in the energy expenditure is probably compensated by an increase in glucose metabolism. Since glucose is the primary source of energy production in the brain, it is reasonable that the system would prefer to utilize the available glucose by reducing its mobilization from one cell to another, by decreasing the G-6-Pase, rather than increase in the activity of hexokinase. However, on long term deprivation since the activity of hexokinase is increased, the decrease in the G-6-Pase activity is reduced.

Long term REM sleep deprivation (E4) showed a tendency for an increase in the activity of LDH, though it was not significant.
Major isoenzyme of LDH in the brain is of H type (Lajtha et al. 1986), which converts pyruvate to lactate. Nevertheless, brain also contains the M type of isoenzyme. Very little increase in the activity of LDH after REM sleep deprivation is difficult to explain specially on the background of previous reports in which infusion of lactate induced REM sleep (Kawakami and Yoshida, 1965) while REM sleep deprivation is known to cause an increase in the levels of lactate (Mendelson et al. 1974). One of the possibilities could be that REM sleep deprivation may not increase the production of lactate but may reduce its metabolism and thereby increase its concentration on deprivation leading to REM pressure or vice versa. It is difficult to comment, from this study, regarding the changes, if any, in the activities of different isoenzymes of LDH.

The activity of G-6-PDH did not show any significant change after long term REM sleep deprivation (E4). G-6-PDH is the enzyme involved in the generation of the reducing factor (NADPH) which is required for the biosynthesis of fatty acids and lipids. The activity of this enzyme is regulated by the levels of glucose-6-phosphate and NADPH. Since REM sleep deprivation probably induces an overall increase in the catabolic processes, it is possible that there may be a decrease in the overall biosynthetic processes. Hence, the activity of G-6-PDH probably did not show any change after REM sleep deprivation. This in turn indicates that the deprivation induced increase in glucose metabolism possibly utilizes the glycolytic pathway.

Thus, this study probably suggests that increased energy expenditure on REM sleep deprivation was induced initially at least by a decrease in the activity of G-6-Pase, which reduced the mobility of glucose from one cell to another, thereby increasing the availability of glucose to be metabolized. As the period of deprivation increased
there was an increase in the hexokinase activity, possibly as a compensatory mechanism, leading to an increase in glycolytic flux. The activities of LDH and G-6-PDH did not show any significant alterations after REM sleep deprivation. Therefore, it may possibly be said that the increase in the glucose metabolism, on REM sleep deprivation, takes place via the aerobic glycolytic pathway. The findings of this study would probably form the basis for further in depth investigation of those enzymes as well as the mechanism for inducing such changes on REM sleep deprivation.

F] EFFECT OF REM SLEEP DEPRIVATION ON ACTIVITY OF 5'-NUCLEOTIDASE:

The significant finding of this study is that REM sleep deprivation (E) caused a decrease in the rat cerebral 5'-nucleotidase activity. The decrease was expressed first in the cerebrum after 96 h REM sleep deprivation. Two days deprivation was not effective in inducing any change in the activity of the enzyme. Results of this study indicated that REM sleep deprivation does affect the metabolism of adenosine. Decrease in the activity of 5'-nucleotidase after 96 hrs deprivation and no change after 48 hrs deprivation is supported by earlier findings that REM sleep deprivation does not cause any alteration in the levels of adenosine after 48 h and REM sleep deprivation affect the A1 receptors of adenosine (Yanick and Radulovacki, 1987). Regarding the mechanism of REM sleep deprivation induced decrease in 5'-nucleotidase activity, the following possibility may be put forward that REM sleep deprivation might induced a decrease directly or indirectly in the activity of 5'-nucleotidase which may be necessary to maintain the levels of adenosine. It may also be possible that REM sleep deprivation may affect the membrane per se and this may cause the alterations in the activity of 5'-nucleotidase which is a membrane bound enzyme. Which
ever possibilities may be true, if at all, at this stage it is difficult to comment on the kinetics of 5'-nucleotidase. This study would form the basis for future study of REM sleep deprivation induced changes in the adenosine metabolism and would help in elucidating the role of adenosine in regulation of sleep and wakefulness.

G) EFFECT OF REM SLEEP DEPRIVATION ON MEMBRANE FLUIDITY:

The significant observation made in this study was that there was a significant change in the fluidity of the microsomal membranes after 24 hrs and 96 hrs REM sleep deprivation. Diphenyl hexatriene (DPH), the fluorescent probe used in this study, has been shown to localize in the fatty acyl side chain region of the lipid bilayer (Lebel and Schatz, 1990). Hence, REM sleep deprivation probably affects the central acyl region fluidity of the microsomal membranes. Alteration in the membrane fluidity are known to be closely linked to variety of factors viz. alteration in the receptor densities (Lebel and Schatz, 1989), alteration in the activities of phospholipases, alterations in the methylation of phospholipids (Lebel and Schatz, 1990) etc.

REM sleep deprivation induced a significant increase in membrane fluidity in the rat brain, may be explained as follows: First, the deprivation might have a direct effect to cause an increase in the receptor densities, which in turn may affect the fluidity of the membrane. Though the mechanism of increase is yet to be investigated, it may be supported by earlier report which have shown that REM sleep deprivation may decrease the densities of NE receptors (Mogilincka et al. 1980, 1986). Second, REM sleep deprivation induced alterations in the activity of muscarinic or adrenergic receptors may induce an changes in the activities of phospholipases especially phospholipase C thereby causing an
alteration in the membrane dynamics (Lebel and Schatz, 1990). Third, REM sleep deprivation may cause an increase in the proposed sleep factors or neuropeptides which may directly or indirectly affect the membrane fluidity. Fourth, the present finding may explain earlier observations that REM sleep deprivation causes a change in the membrane bound enzymes viz. AChE, Na-K ATPase, 5'-Nucleotidase etc. which may be cause change in the fluidity of the membrane or vice versa.

One of the interesting observations is that short term as well as long term REM sleep deprivation caused a significant increase in the membrane fluidity in the cerebrum and the brain stem, the cerebellum remained unaffected even after four days of REM sleep deprivation. Increase in the cerebrum fits well with the earlier findings that after REM sleep deprivation, there is a decrease in the cortical β-adrenergic receptor sites (Mogilinicka et al. 1986). It may also be possible that the activity of Na-K ATPase which increases in cerebrum on REM sleep deprivation (Gulyani and Mallick, 1993) may be due to the alteration in the membrane fluidity and Na-K ATPase, an integral membrane protein is very sensitive to the alteration in the membrane dynamics. The increase in membrane fluidity in the brain stem may have relevance to the fact that brain stem is the region known to be the REM generator and hence, deprivation induced alteration in the membrane fluidity may influence the firing rate of the brain stem neurons (Mallick et al. 1989, 1991). Changes in the behavior of the E rats due to deprivation has been reported earlier (Vogel, 1975). It is difficult to comment from this study if those behavioral changes have any relevance to an increase in the brain membrane fluidity. The finding of this study would probably form the basis for further investigation regarding changes in the turnover of phospholipids, alteration in the fatty acid
The levels of phospholipids did not show any significant change after long term (four days) and short term (two days) REM sleep deprivation. Phospholipids are the most important components of the brain and are involved in varied functions. As REM sleep deprivation altered the membrane fluidity it was hypothesized that there may be alteration in the levels of phospholipids. It may be possible that there may be alterations in the turnover of phospholipids taking place with the levels remaining unchanged or it may be that there is an alteration taking place in the fatty acid components of the phospholipid. Further detailed investigation is needed to understand the effect of REM sleep deprivation on the membranes constituents.