CHAPTER I

INTRODUCTION AND REVIEW OF LITERATURE

Alcoholism is an age old practice and its effects on the liver and other major organs have always been cause for concern. Current research shows that all the major systems including the immune system is affected by alcohol intoxication. One of the most devastating effects of alcohol is on the central nervous system. Each part of the brain is highly vulnerable to the presence of alcohol and each of these parts respond differentially to alcohol. It can be observed at each level, be it the receptor level, blood flow level or transmitter level. Alcohol acts through carrier systems and second messengers and is known to produce membrane damage through increased lipid peroxidation. It acts on different enzyme systems. Reduction in glutathione peroxidase activity was observed in cerebral cortex, cerebellum and brain stem of rat brain. Vitamin E acts independently and in association with glutathione system to protect against alcohol induced damage (Dietrich et al 1989, Marcus et al 1993).
Alcohol may function as a selective N-Methyl D-aspartate (NMDA) subtype of glutamate receptor. The effect of alcohol can be reversed by high concentration of glycine. Membrane preparation studies indicate that alcohol may act by increasing frequency of ion channel opening. The number of NMDA receptors increased in many brain areas. This up regulation of receptors may be associated with withdrawal seizures. (Meller et al 1993. Tabakoff et al 1993). Autogenization of NMDA receptors could account for significant portion of the amnestic and sedative actions of alcohol (Lovinger et al 1989). Continuous exposure to alcohol was found to cause significant deficits in the body and brain weights. The concentration of total ganglioside, on the other hand, in whole brain, cerebrum, cerebellum and brain stem showed an increase following exposure to alcohol. The activities of sialidase, beta-galactosidase, beta-glucosidase and beta-hexoxaminidase which are likely to be involved in the catabolism of gangliosides showed reduction. Alcohol was also found to alter the proportions of individual gangliosides and changes were found to be region specific. Alcohol induced alterations were reversed to some extent upon abstinence. (Prasad 1993, Klemm et al 1979).
The direct interaction of alcohol molecule may involve GABA$_a$ receptor (Suzdak et al 1987). Experiments have revealed that alcohol inhibited NMDA receptor - mediated synaptic currents without potentiation of GABA (A) events or attenuation of GABA (B) - mediated fading of GABA (A) synaptic currents. The effects of alcohol on long term changes in synaptic strength on the rat hippocampus formation are primarily due to an action at the NMDA receptor - channel complex (Morrisett et al 1993).

Alcohol decreased the sialic acid content of the brain (Klemm et al 1979). Alcohol has been shown to affect the GABA system by suppressing GABA catabolism (acute) or synthesis (chronic) or by altering the affinity of GABA receptor. Chronic alcohol can alter the expression of POMC gene in the hypothalamus (Kulonen et al 1983, Angelogianni et al 1993). An adduct of stable acetaldehyde enkaphalin in dilute buffer led to alteration of biological and immunological activities. Acetaldehyde adduct formation destroys the binding of two major endogeneous enkaphalins at $\varepsilon$ and $\mu$ central opiate receptors. There is no barrier between the brain tissue and CSF. Alcohol ingestion may effect the central endocrine mechanism through alterations on activity of endogeneous cerebral opioids (Lightman et al 1980). Acute and chronic alcohol
increased the levels of $\beta$-endorphin in plasma and CSF of humans (Thiagarajan et al 1989).

Cytosolic Ca$^{2+}$ functions as a main regulator in numerous cellular processes including those implicated in synthesis, release and responsiveness of hormones. Ca$^{2+}$ levels in the peripheral blood mononuclear cells of alcoholic patients was significantly higher. Changes in Ca$^{2+}$ may be relevant to increased excitability of the CNS (Nagy et al 1993).

A general reduction on membrane associated protein kinase C was observed in vitro. The differential effect in the CA1 region of the hippocampus may be a reflection of disruption in the normal regulation of protein kinase C activity in this area. (Kruger et al 1993). Acute ethanol administration resulted in time and dose dependent decreases in the levels of inositol monophosphates (IP-1) in cerebrum and cerebellum. Decreased IP-1 levels correlated well with decrease in the levels of inositol -1,4, 5-triphosphate and the increase in blood alcohol concentration (Lin et al 1993). Decrement of cholinergic function have been implicated as a cause of deterioration of memory in chronic and acute alcohol abuse (Beracochea et al 1986). An interaction between alcohol and nitcotine has been suggested through cholinergic nicotinic receptors (Dar et al 1993).
Alcohol has an effect on all the three monamine transmitters NA, DA and 5 HT (Nutt et al 1986). Dopamine is thought to be involved in the reinforcing actions of alcohol. Systemic and locally infused alcohol releases dopamine in the nucleus accumbens. Alcohol withdrawal is associated with reduced release in this pathway (Rossetti et al 1992). In low doses, alcohol increases utilization of dopamine (Lai et al 1979). Acute administration of low doses of alcohol increases synthesis of DA (Carleson et al 1973, Hunt et al 1974, Di Chiara et al 1988). Acute and chronic ethanol significantly down regulates the neurotensin receptor system indicating that the action of alcohol may be partly mediated by neurotensinergic systems (Campbell et al 1993).

Alcohol alters normal endocrine function through activation of mixed function oxidases, alteration in sleep patterns or a direct effect on hormone formation and release (Lightman et al 1980). Ethanol decreases GHRH followed by a concomittant decrease of GH (Dees et al 1990, Badger et al 1993, Fernstorm et al 1995).

Thyroid hormones which play a major role in metabolism, growth and development are also seen to be affected by alcohol abuse. Acute intoxication results in transitory hyperthyroidism while chronic intake of
alcohol decreases hormone levels. Parathormone levels increased and Vitamin D metabolites decreased following alcohol consumption (Zima 1993). TRH reduces the sedative effects of ethanol (Cott 1976). TRH antagonises the hypnotic, hypothermic and motor impairing properties of ethanol (Nemeroff et al 1984). In the CNS, the activity of type II-5 deiodinase which catalyses deiodination of T_3 and T_4 was lower in behaviourally dependent rats and controlled drinkers as compared to the alcohol naive controls in the frontal cortex, parieto occipital cortex, hippocampus and striatum but not in the cerebellum or pituitary. T_4 levels were higher in the areas of the CNS in the groups exposed to alcohol whereas T_3 concentrations were normal. The activity of Type III 5 - deiodinase (which catalyses the further deiodination of T_3) did not change. Abstinence for three months led to the normal activity of type II 5, deiodinase. Type III 5 - deiodinase activity was inhibited possibly to maintain physiological concentration of T_3 during abstinence. The tissue levels of T_3 were normal in the areas of CNS and T_4 level elevated (Baumgartner et al 1994).

Decreased TSH response to TRH has been observed in subjects with a history of alcoholism. This cannot be explained on the basis of down regulation of TRH receptors because then Prl response
would also be diminished. Even in abstinent alcoholics there is a lower TSH response to TRH (Garbutt et al 1991(b)). During withdrawal, in chronic alcoholics, abstinence decreases free $T_4$ and free $T_3$ ($fT_4$ and $fT_3$) levels whereas the respective protein bound fractions are normal. $T_3$, $T_4$ and TBG increased during abstinence, reverse $T_3$ ($rT_3$) concentration decreased but not significantly. TSH values did not show any change. Low levels of serum $fT_3$ and $fT_4$ during withdrawal has been attributed to increased tissue uptake (Baumgartner et al 1994).

Chronic alcohol leads to decreased levels of $T_4$, $fT_4$, $T_3$, $rT_3$ and basal TSH Secretion (Mason et al 1988). Acute alcohol treatment also reduces serum $T_3$, $T_4$ and free $T_4$ levels (Orrego et al 1979). In the liver the concentration of $T_3$ and $T_4$ were lower in behaviourally dependent animals in the controlled drinkers after 3 months of abstinence (Baumgartner et al 1994). Thyromegally and reduced secretion of $T_3$ as well as an increased conversion of secreted $T_4$ of $rT_3$ as opposed to $T_3$ was reported in men with advanced alcoholic liver disease (Orrego et al 1979).
ALCOHOL AND GENERAL METABOLISM:

Alcohol increases heart rate, skin temperature and blood pressure. Alcohol causes peripheral vascodilation with increased cardiac output, tachycardia and increased blood pressure. (Higgins et al 1993, Zima 1993) Moderate drinkers have an increased HDL-cholesterol with diminishing risk of coronary heart diseases. Acute alcohol intake causes an increased level of triglycerides without changes in HDL-cholesterol level (Zima 1993). Acute administration of moderate dose of alcohol caused a decrease in glucose uptake in normal men. Plasma insulin level was also elevated (Jarvinen et al 1993). Carbohydrate metabolism in chronic alcoholics is characterised by insulin resistance and impaired glucose tolerance (Andersen et al 1983). Pure alcohol is a mild stimulant of acid secretion whereas at higher concentrations it has either no effect or a mildly inhibitory one. Alcoholic beverages with low ethanol content are strong stimulants of gastric secretion and gastrin release. Beverages with higher ethanol content do not stimulate gastrin release (Chari et al 1993). Chronic alcoholics have increased post prandial pancreatic enzyme secretion. The post prandial hypersecretion of enzymes in alcoholics is not related to increased plasma levels of CCK or gastrin. It could be due to the impaired release of pancreatic polypeptide which
Chronic EtOH consumption (Brain protein synthesis)

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Reduced Protein synthesis by soluble and membrane bound ribosomes

↓

Selective for certain regions like Hippocampus/Amygdala;

Cerebellum

↓

Decreased neuronal growth and maturation

↓

Chronic alcohol intake and its effect on brain protein synthesis and neuronal development

(Michaelis 1990)
participates in increased pancreatic enzyme secretion (Hajnal et al 1993). Adaption of gastric mucosa to chronic alcohol administration is associated with cell proliferation, increased expression of mucosal EGF, TGF and EGFR (Epidermal Growth Factor Receptor) (Tarnawski 1992). Liver glycogen decreases and plasma glucose increased following alcohol exposure (Simm et al 1990). Alcohol abuse may result in central diabetes insipidus (Zima 1993). Obesity is related to elevations of serum alanine ammno transferase which is brought about by alcohol. Insulin is thought to play a role in this relationship (Fagerberg et al 1993).

Chronic alcohol treatment results in disturbed Vitamin D metabolism, hypomagnesemia and skeletal changes (Turner et al 1988). Bone marrow cellularity diminishes with subsequent reduction in erythropoiesis, thrombopoiesis and leukopoiesis. Alcohol causes sideropenic and megaloblastic anemia. There are two forms of alcohol muscle injury, the acute myonecrosis and inflammatory reaction and chronic one with muscle weakness and atrophy. Alcohol can also cause osteoporosis (Zima 1993).

Immunosuppression observed in chronic alcohol users is caused by multiple factors including the amount of alcohol induced nutritional deficiencies. Spleen cell number, interleukin -2 and tumour necrosis
factor (TNF) secretion were independent of the diet consumed, but is affected by consumption of alcohol. Body and spleen weights and interferon gamma secretion were modulated by alcohol as well as by diet. Nutritional composition of the diets consumed during concurrent administration of alcohol modulates the immunotoxic effects. Alcohol reduced number of lymphocytes, reduces phagocytosis by macrophages and diminishes the activity of NK Cells (Watzl et al 1993, Zima 1993). Alcohol has been observed to cause a higher frequency of numerical and structural aberrations (Badr et al 1982).

The major route for alcohol metabolism requires a two step oxidative process. In the first step it is catalysed by ADH (alcohol dehydrogenase) and in the latter by ALDH (aldehyde dehydrogenase) (Guan et al 1989). The primary pathway for the metabolic breakdown of ethanol is thought to be the conversion of alcohol to aldehyde by alcohol dehydrogenase and then to acetic acid following oxidation of the latter by aldehyde dehydrogenase (Idanpaan - Heikkila et al 1972, Brien et al 1983). Acetate generated after alcohol metabolism potentiate some of the anesthetic actions of alcohol through adenosine (Carmichael et al 1992).
At low concentrations alcohol is metabolised largely by alcohol dehydrogenase to acetaldehyde while at higher concentrations a microsomal ethanol oxidising system (MEOS) is involved namely P450 1IE1 which also generates free radical (FR) species. (Teare et al 1993). An increased formation of FR (free radicals) in tissues would increase their oxidative stress and may increase their susceptibility for developing chemically induced cancers. FR and some FR products can rapidly react with biological materials like lipids, proteins and nucleic acids, forming toxic products. Free radicals and their products are cancer promoters. Diets supplemented with high levels of Vitamin E which inhibits alcohol induced free radical activity and the formation of FR products suppress the promotion of cancer by alcohol (Eskelson et al 1993). Brain sialoglyco conjugates are hydrolysed after alcohol treatment (Cherian et al 1989).

**Alcohol and the Female System**

Studies of alcohol in female is lesser than those found in males. It may be due to the difficulty in correlating with the female cycle. Alcohol infact acts at all levels in the female, be it prepubertal, pre-menopausal or post menopausal effect of alcohol has been observed from the hypothalmic level to the mammary gland.
MATERNAL ALCOHOL INTOXICATION

Direct inhibition of protein synthesis in fetal organs

eg: Brain and liver

Ribosomes and Enzyme activities decrease

Decreased organ development

(Weiner et al 1981)
Umbilical Blood Vessel spasms

↓

Brain and other organ Hypoxia

↓

Glutamate release

↓

Increased excitability - NMDA receptor activation

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Ca²⁺ entry

↓

Protease activation and lipid peroxidation

↓

Cell death

(Michaelis 1990)
Alcohol treated rats showed a significant increase in the hypothalamic content of LHRH with a significant decrease in the serum concentration of LH but not FSH. Chronic prepubertal alcohol administration alters the concentration of specific hypothalamic and pituitary hormones involved in the female pubertal process (Dees et al 1990). Alcohol has been shown to suppress LHRH/LH secretion through CRF induced EOP and probably β-endorphin release (Rivier et al 1984, 1992). Spontaneous release of LH has been observed following alcohol treatment suggesting that alcohol inhibits ovulation by affecting the hypothalamic pituitary axis (Wilson et al 1978). Lower doses of alcohol and sucrose administration were followed by a significant decline in LH. However, FSH levels did not change after sucrose administration in recently ovariectomised monkeys but was significantly suppressed after being given a higher dose. In chronically ovariectomised monkeys FSH decreased significantly after administration of sucrose and different doses of alcohol (Mello et al 1986, 1987, 1989). Gonadotropin stimulation during alcohol intake potentiates the action of LH in women (Mendelson et al 1988, Wimalasena et al 1993)).

Alcohol was observed to disrupt the normal estrous cycles in females as seen through vaginal epithelial changes (Eskay et al 1981),
Vaginal blood flow decreases as alcohol level in serum increases (Fuchs et al 1963). Female rats following a seven week alcohol diet showed atrophy of uteri, fallopian tubules and ovary. Ovary showed absence of well developed follicles, corpora lutea, corpora haemorrhagica and secretory granulosa cells (Fuchs et al 1980). Ovarian LDH decreased after IP injection of alcohol in pregnant mice (Chernoff et al 1977). Inhibition of ovulation and significant reduction of estrogen and progesterone levels have been reported in rats after alcohol administration (Wilson et al 1978). Alcohol decreases gonadotrophin induced protein synthesis in vivo and in vitro (Poso et al 1981).

Alcohol consumption was seen to reduce ovarian weight, corpora lutea were lost and plasma concentratin of estrogen, progesterone and gonadotropins were low (Van Thiel et al 1978 a). Secondary amenorrhea was observed in female alcoholics (Zima 1993). Alcohol at a concentration of 40 mM was reported to enhance estrogen while inhibiting estrogen sulfate transport. Lesser estrogen was transported as free estrone in the presence of alcohol in controls (Martin et al 1982, Valimaki et al 1983).

In pre-menopausal female alcoholics, alcohol consumption increases the frequency of menstrual disturbances, abortions and miscarriages
while infertility is not frequent. Acute alcohol intoxication has only minor effects on pituitary gonadotropin hormones in pre menopausal women while chronic alcohol abuse led to reduced concentration of sulphated steroids and these changes may be seen before severe liver dysfunction has occurred. In women liver dysfunction lead to earlier occurrence of menopause in comparison with normal controls. In post menopausal women with alcoholic liver disease, the main disturbance of sex hormone metabolism consists of elevated estrone and sex steroid binding globulin concentration, while serum concentration of steroid sulfates and 5αHDT are reduced. The presence of high affinity low capacity, specific estrogen receptor has been confirmed (Becker 1993).

Prepubertal administration of 5% alcohol in female mice causes delay in vaginal opening (Bo et al 1983). Most epidemiologic studies of the relationship between alcohol consumption and breast cancer risk show that persons who consume moderate amount of alcohol are at 40 - 100 % greater risk of breast cancer than those who do not consume alcohol. Studies in women (21-40 yrs) with regular menstrual cycles show that alcohol was associated with increases in several hormones. Plasma dehydro epiandrosterone sulphate levels were 7 % higher in the follicular phase. In the periovular phase there is an increase in plasma estrone
plasma estrogen and urinary estrogen levels. In the luteal phase urinary levels of estriol rose. Increase in total estradiol levels in periovulatory phase suggest elevated absolute amounts of bioavailable estradiol. Total estrogen and bioavailable estrogen increase with alcohol consumption. This should be an explanatory mechanism for breast cancer in premenopausal women (Reichman et al 1993).

Alcohol consumption causes an increased risk for ovarian cancer. The association of very low levels of alcohol intake with breast cancer risk may be due to confounding whereas three or more glasses of alcoholic beverages daily appears to be genuinely increase breast cancer risk in pre and post menopausal women (Gapstur et al 1988, Polychronopoulou et al 1993, Katsouyanni et al 1994). Significant multiplicative interaction between non contraceptive estrogen use and alcoholic intake has been observed. No association between alcohol and breast cancer was observed among never users of estrogen in post menopausal women (Gavaler et al 1992).

Alcohol consumption can alter mammary gland structure, early stages of lactation even when adequate levels of dietary protein are maintained (Sanchis et al 1986, Steven et al 1989). Intraperitoneal
Injection of alcohol to Swiss mice decreased liver glycogen, increased plasma glucose, uterine glucose, glucose - 6 - PO4, fructose - 6 - PO4 and citrate (Simm et al 1990).

**Alcohol and the Male System:**

The effect of alcohol on the male system is one of the major areas that have been observed with keen interest. Alcohol has been observed to affect the male system at all levels (Ida et al 1992). Alcohol reduces the serum LH and testosterone levels which was increased by the administration of LH or norepinephrine. The reduction on androgen receptors, serum LH and testosterone, but enhancement of these by treatment with neurohormone and neurotransmitter suggests that ethanol exerts an adverse effect on the hypothalamic pituitary unit and the neurotransmitter hypothalamus hormone relationship resulting in impairment of the androgen induced sexual events (Kakihana et al 1980; Chung et al 1989, Blanchard et al 1994).

Multiple administration of ethanol stimulates deficit of serum testosterone, androgen receptors in medial basal hypothalamus and pituitary body that probably results in separation of negative feedback mechanism between the gonads and pituitary body. Increase in
specific binding of estrogen to nuclear receptor in the preoptic area (POA) might appear to explain the feminization of alcohol treated rats (Babichev et al 1989).

Serum LH and testosterone levels were significantly decreased in alcohol fed rats (Badr et al 1979, Chung et al 1989). Serum FSH levels were also reduced. The ratios of pituitary LH and FSH to their serum levels were clearly increased after alcohol exposure, pituitary prolactin decreased, testicular LH receptors reduced, FSH receptors increased slightly. No change was observed in the levels of testicular prolactin, GnRH or pituitary GnRH receptors. Alcohol was found to decrease FSH-β and increase LH-β mRNA and to have no effect on the common α subunit and prolactin mRNAs in the rats given chronic alcohol (Salonen et al 1990, Adams et al 1991, Salonen et al 1992). An exaggerated response of LH to LHRH was also evident after one week of ethanol treatment. Plasma estrogen levels increased. Elevation in plasma prolactin and estradiol could be the contributing factor to the maintenance of hypogonadism (Esquifino et al 1989).
Acute alcohol decreased levels of $1\beta E$ and increased serum levels of the peptide for one hour after its injection (Tsong et al 1982, Adams et al 1991). Ethanol depletes gonadotrophin binding sites in the testes. Rats given a large dosage of alcohol for seven days have been shown to have a 30-35% decrease in testicular gonadotrophin receptor concentration (Bhalla et al 1979). Alcohol reduces androgen receptor numbers (Bhalla et al 1979, Eagon et al 1985). Alcohol also increases the number of estrogen receptors and deletes estrogen metabolising tissues in such tissues (Eagon et al 1985, Andersson et al 1986).

In humans also there is a decrease in plasma testosterone after chronic alcohol abuse. Acute and moderate doses do not cause any change (Mendelson et al 1988). Reduced concentration of plasma, total testosterone and free testosterone have been found in patients with cirrhosis of the liver. Alcoholic men have decreased basal testosterone and elevated LH and FSH (Mowat et al 1976, Van Thiel et al 1978b). Alcohol was found to decrease FSH-$\beta$ and increase LH-$\beta$ mRNA. No effect was observed on the common subunit and Prl mRNA (Salonen et al 1992).

Higher dose of alcohol inhibits testicular testosterone synthesis. It also causes an elevation in the intratesticular pregnenolone to progestrone
Corticosterone also seems to play a role in the inhibition of steroidogenesis (Orpana et al. 1990). Major microsomal enzymes involved in the biosynthesis of testosterone like 3-beta hydroxysteroid dehydrogenase and steroidogenease mixed function oxidases were markedly inhibited in a dose and duration dependent manner. The terminal enzyme 17-beta hydroxy steroid dehydrogenase unaffected by ethanol treatment except at higher doses (Anderson et al. 1983, Singh et al. 1991). Alcohol and acetaldehyde inhibits testicular 17-β hydroxysteroid oxidoreductase, an enzyme necessary for conversion of androstenedione to testosterone, (Cicero et al. 1981). Alcohol reduces testicular NAD levels (VanThiel et al. 1980). Following alcohol administration, ADH activity in testicular interstitial tissue was increased suggesting that ADH is involved in alcohol metabolism in the testis (Shirai et al. 1992). The content of testis cAMP, leu-enkaphalin and β-endorphin is unchanged, thus excluding the opioid system from the mediation of effects of alcohol (Kokha et al. 1989). Opioid antagonists reduce alcohol intake (Samson et al. 1985).

Lower doses of alcohol did not result in testicular atrophy but at higher doses, the testes contained smaller semineferous tubules, decreased the numbers of total cells without causing degeneration in
the spermatids. In the peritubular walls of the semineferous tubules, there were curvature irregularities, unfolding of the basement membrane and lamination of the lamina densa as well as hyperplasia of collagen fibres in the tunica propria (Shirai et al., 1992).

Lower doses of alcohol did not cause significant differences in body, testes or prostate weights. Seminal vesicle weights decreased significantly in the alcohol treated rats (Van Thiel et al. 1975, 1979, 1980, Salonen et al. 1990). Acute alcohol increased testicular interstitial fluid 1 - βE and decreased testicular interstitial fluid volume. (Adams et al. 1991). Alcohol consumption produces damage to the testicular germinal epithelium and subsequently fewer mature spermatoza are developed (Arlit et al. 1971). Sertoli cells may be a target for gonadal toxicity. Alcohol depletes ATP reserves in Sertoli cells and alter intracellular Ca$^{2+}$ homeostasis (Farghalli et al. 1993). Chronic alcoholism causes testicular atrophy. This could be due to the conversion of retinol to retinal which is mediated by the zinc metal enzyme alcohol dehydrogenase. (Mc Clain et al., 1979). A decrease in sperm motility is thought to be secondary to an increased amount of mucus in the semen as a result of irritant effect of alcohol on prostrate gland (Moinar et al. 1973). In humans and animals, observations have shown that alcohol consumption
produces significant changes in spermatozoal morphology like break-
age of sperm head, distension of midsection and curling of its tail. Adult
male rats showed significant atrophy of seminal vesicles, prostrate and
testes (Van Thiel et al., 1980). Chronic alcohol treatment caused re-
duction in testicular weights, epididymal sperm content and reduced
sperm motility. Epididymal weights were unaffected. Disorganisation of
spermatogenesis, germ cell degeneration, decreased tubular luminal
diameter and vacuolation of Sertoli cells were seen after alcohol
treatment (Anderson et al., 1989). One of the major effects of alcohol is
feminization. As a consequence of alcohol induced increment of
hepatic aromatase activity the conversion of androgens (testicular and
adrenal origin) to estrogen is increased in the liver (Gordon et al 1979,
1980).

**Fetal Alcohol Syndrome**

The effect of alcohol on prenatally exposed individuals and also
consumption of alcohol by both adult male and female has been an
interesting area of study.

Fetal Alcohol Syndrome (FAS) is recognised as a pattern of major
and minor malformation, growth deficiency and developmental
disability caused by heavy alcohol exposure in utero. The main manifestation of FAS is growth deficiency, which is usually of prenatal onset and continues postnatally, a particular pattern of malformations, and malformations of CNS often manifested as microcephaly, mental retardation, motor problems, hyperactivity etc., The frequency of skeletal anomalies, heart defects and other abnormalities are also observed (Clarren et al 1978, Streissguth et al 1980, Frye et al 1981, and Gilliam et al 1985).

Some of the effects of prenatal alcohol exposure appear to be dose specific and are observable beyond a certain threshold. The procedures necessary to elicit alcohol-related differences vary depending on the amount of exposure and the severity of the effects anticipated. Behavioural effects of prenatal alcohol exposure appear to be detectable at exposure levels below those necessary to detect growth and morphological effects.

Prenatal alcohol exposure decreased the cerebral content of acetylcholine in fetuses aged 18 and 21 days and neonatals aged 3 and 6 days, but had no effect on the precursor of acetylcholine, choline and increased cerebral acetylcholinesterase. Increase in isolation - induced activity displayed by alcohol-treated pups at 22 days is due in part to a
delay in the functional development of a cholinergic system involved in the inhibition of activity (Rawat 1977).

In utero alcohol exposure leads to a decrement in the ability of the animal to inhibit a prepotent response. It has been suggested that the hippocampus would be a likely place to locate the physiological sequelae of this syndrome (Abel 1978, Abel 1981, West et al 1983, and Abel 1989).

Alcohol is transferred across the placenta and reaches an approximate equilibrium between the maternal blood level and the fetal blood and tissue. The primary pathway for the metabolic breakdown of alcohol is thought to be the conversion of alcohol to acetaldehyde by the action of alcohol dehydrogenase and then to acetic acid, following oxidation of the latter by aldehyde dehydrogenase. The rate of ethanol formation from fetal tissues especially the amniotic fluid is considerably slower than that for the maternal blood levels. (Idanpaan-Heikkila et al 1972, Wattman et al 1972, Keasaniemi et al 1975 and Brien et al 1983). This slow rate of elimination has been attributed to the lack of fetal alcohol dehydrogenase activity during the early gestation (Raiha et al 1967, Pennington et al 1983). Therefore it has been suggested that ethanol dissolution in the amniotic fluid may lead to a prolonged exposure of the fetus to elevated levels of this
alcohol (Brien et al 1983). Both alcohol by itself and acetaldehyde have been shown to have direct inhibitory effects on DNA synthesis and protein formation in embryonic tissue (Dreosti et al 1981). Introduction of increasing concentration of alcohol to cultures of rat embryos from day 10 of gestation produced dose-related retardation in growth and development of treated embryos and decrease in total DNA and protein content of embryonic tissues (Brown et al 1979). The two main features of the brain abnormalities are (1) the neuronal populations most frequently affected by prenatal or early postnatal alcohol exposure are those that have already achieved a certain level of maturity rather than those that are undergoing cell division prior to migration, (2) the brain dysmorphologies involve axonal growth changes and altered migration patterns of neurons rather than elimination of neuroblasts. Some investigators have ascribed the toxic events and metabolic changes that follow exposure of cells to alcohol to physical and chemical changes within plasma or cellular organelle membrane structure and function (Freund et al 1976, Rottenberg et al - 1980). The best documented activity of the neutral alcohols is their physical interaction with the plasma membraneous organelles (Harris et al 1981, Michaelis et al 1983 a: Goldstein 1984). Alcohol has been shown to inhibit directly the
synthesis of DNA in isolated, mitogen stimulated spleen cells in culture (Freund et al. 1976). Alcohol is said to induce cytotoxicity through changes in the lipid organisation of cell membranes possibly including direct effects on membrane proteins (Li et al. 1980).

Alcohol exposure causes retardation in the development of substantia nigra and pars compacta neurons especially in their dendritic growth and branching leading to abnormal motor function (Shetty et al. 1993). The membrane disordering effect of alcohol is in some respects similar to the increases in lipid fatty acid chain motion produced by elevations in ambient temperature. Adaption of brain neurons, erythrocytes and liver cells of animals exposed to chronic ethanol treatment involves a greater resistance of the cell membranes to the increase in lipid motion produced by alcohol as well as changes in the temperatures that induce phase transitions in these membranes (Chin et al. - 1977a, 1978). The decrease observed in both body and brain weights of infants born to alcoholic mothers and of experimental animals exposed to alcohol during gestation suggest that the cellular protein synthetic machinery might be quite sensitive to the presence of alcohol within the fetal brain. Alcohol exposure leads to a reduction in cell number, perhaps as a result of decreased cell division brought about by Zn 2+ deficiency and
lower thymidine kinase activity. The possibility that the reduced brain size and actual loss of cells may be due to interference with protein synthesis has not received much attention in FAS. Alteration of growth and development is consequence of alcohol exposure (Adickes et al 1990). The cerebellar neuronal changes in rat pups following gestational exposure to alcohol may be the result of an alcohol - induced hypothyroid state (Kornguth et al 1979). Another possible indirect mechanism for alcohol toxicity - is that neuronal cell loss caused by circulatory changes leading to hypoxic cell damage within the brain. Following alcohol administration to a mother monkey, a rapid collapse of the umbilical blood vessels was observed within minutes of a bolus injection followed by a fall in fetal blood PO2 and pH and a rise in PCO2 (Mukherjee et al 1982).

In vitro studies in humans have demonstrated that alcohol produced constriction of human umbilical arteries and veins. The alcohol - induced vasoconstriction was dependent on the dose of alcohol used and apparently not the result of cholinergic, adrenergic, serotonergic or prostaglandin related activity (Altura et al 1983). Susceptibility of neurons to hypoxia may be due to the action of glutamate on excitatory receptors of the NMDA variety (Simon et al 1984).
Hypoxic cell damage may not be so much a reflection of inhibitory cell loss or excessive excitatory inputs as a shift in the balance between excitation and inhibition. Thus balance could shift with exposure to hypoxia because of alterations in the maintenance of ionic gradients across the neuronal membrane. A change in the equilibrium potential for $\text{Cl}^-$ may affect the responsiveness of neurons to GABA. Similarly a change in the neuronal membrane system for transporting $\text{Ca}^{2+}$ or $\text{Na}^+$, the primary ions involved in the initiation of excitatory amino acid - induced depolarization may increase susceptibility of these neurons to the damage produced by L-glutamate and its analogs (Takeuchi et al 1973). Of these ions, the accumulation of intracellular $\text{Ca}^{2+}$ is considered to be the event most closely associated with cytotoxicity (Donaldson et al 1983). Maternal alcoholism could result in intermittent, transient episodes of ischemia and hypoxia for the fetal brain and may eventually lead to a compromised capacity of the nerve cells to provide adequate regulation of intracellular $\text{Ca}^{2+}$ levels. The dissolution of alcohol within the neuronal membrane as well as the hypoxia produced by its effect on the cerebral vasculature could alter the neuronal plasma membrane such that it no longer provides an adequate barrier to $\text{Ca}^{2+}$ or an efficient system for $\text{Ca}^{2+}$ extrusion. It has been concluded that
excessive cytosolic Ca$^{2+}$ is quite likely to be the triggering mechanism for ischemic cell injury in these highly vulnerable cells (Donaldson et al 1983).

There is considerable evidence to indicate that alcohol significantly alters the activity and responsiveness of many neuroendocrine systems. Studies involving chronic or acute administration of alcohol both in humans and animals have demonstrated alterations in the secretory function of the gonads, the adrenal cortex and medulla and the thyroid as well as on the secretion of GH, prolactin and vasopressin. Data indicate that alcohol can act directly on the endocrine glands themselves as well as (or in addition to) have an action on the central aspect of hypothalamic pituitary functions (Van Thiel 1980, Cicero 1981, Morgan 1982). The majority of studies performed to date both in clinical settings with animal models of alcoholism have been done in males, possibly in order to avoid complications resulting from cyclic changes associated with estrus or menstrual cycles. There has been little investigation of the extent to which alcoholic influence on neuroendocrine functions in females and virtually no examination of the effects of alcohol on hormonal function during pregnancy. Results from males, however, suggest that a variety of endocrine/neuroendocrine disturbances probably also occur in
females. Thus alcohol induced changes in endocrine functions during pregnancy could have an effect on the female's ability to maintain a successful pregnancy, on the hormonal interactions between maternal and fetal systems and on the orderly development of fetal endocrine function in utero, thus contributing in a number of ways to the etiology of FAS.

Basal levels of $T_3$ were found to be lower in fetal sheep exposed to alcohol. Responses to TRH in terms of $T_3$ and $T_4$ levels were also reduced. TSH responses to TRH injections were also reduced (Rose et al 1981). Reduced serum thyroxine levels were observed on postnatal day 11 in prenatally alcohol exposed rats (Kornguth et al 1979, Hannigan et al 1990). Neonatal injections of alcohol also resulted in increased hypothalamic / serum and pituitary/serum ratios compared to those in control animals (Slebodzinski 1979). Action of alcohol on blood-brain barrier increases the permeability of the barrier for thyroxine. Impaired thyroid function in the neonates could result in a variety of metabolic disturbances that would affect normal development.

Growth hormone: Although some studies indicate decreased levels of GH secretion (Thadani et al 1979), GH responses in children with
FAS have been shown to be normal (to glucose, insulin, arginine, fasting and exercise Barry et al 1975).

Sexual differentiation, maturation and behaviour is seen to be affected by prenatal alcohol exposure in the offspring. Although delayed vaginal opening has been observed in mice exposed to alcohol prenatally it has been suggested that it could be due to undernutrition (Boggan 1979, Ward et al 1994). In male rats, demasculinizing effects have been observed in prenatally exposed rats. Dihydrotestosterone levels in the brain were significantly reduced in newborn alcohol exposed male rats. Both brain and plasma levels of T were also slightly reduced but testicular weight did not show any change (Kakihana et al 1980, McGivern et al 1992). Decreased anogenital distance was observed both in males and females as well as earlier puberty in males after prenatal exposure to alcohol. Penile reflexes were significantly decreased and fewer animals tended to show appropriate sex behaviour in adulthood (Chen et al 1979). This suggests a feminization of alcohol exposed males. Alcohol induced disruption of perinatal androgen status may be responsible for the observed feminization of males either through a direct action of alcohol on fetal testes or through effects of alcohol on T metabolism or utilization (McGivern et al 1984).
Alterations in LH secretion has also been observed following prenatal alcohol exposure. Afternoon LH surges were significantly depressed in alcohol exposed females and there was an overall decrease on secretory pulse frequency. Suppression in plasma LH levels with a concomitant increase in plasma prolactin was observed in pre pubertal animals. Increase prolactin may modify hypothalamic dopamine turnover, thus influencing LH secretion (Guerri et al. 1984).

The suppression of perinatal testosterone surge in male rats exposed to alcohol in vitro and the associated long term demasculinizing effects of prenatal alcohol exposure might be the result of reduced testicular steroidogenic enzyme activity in the perinatal animal. The activity of 17α-hydroxylase was significantly reduced (Kelce et al., 1989, 1990). Alcohol administered daily from day 15 post conception resulted in elevated testosterone (T) levels on day 18 in male and female.

In alcohol treated female, the onset of regular estrous cycle was significantly delayed (Dahlgren et al. 1989). Liquid ethanol with testosterone propionate decreased birth weight, whereas without testosterone propionate the weight did not increase suggesting an inhibition of fetal growth produced by a synergism between activation of the hypothalamic
pituitary adrenal axis and elevated androgen levels (Mc Givern 1989). Acute exposure of mice to alcohol during post implantation pregnancy causes alterations in several glycolytic intermediates in the uterus. Liver glycogen decreased, plasma glucose increased and uterine glucose, glucose 6-phosphate, fructose-6-phosphate and citrate increased (Simm et al 1990). Fetal ethanol exposure may influence gonadal development but not gonadal hormone dependent behaviour. Such animals do not exhibit masculinization of play behaviour (Blanchard et al 1994). Sexual differentiation, maturation and subsequently behaviour of offspring are affected by alcohol exposure. There is a delayed vaginal opening in female fetuses exposed to alcohol (Boggan et al 1979).

In utero alcohol exposure during day 12 of gestation through birth has no apparent morphological effect on the testes of Day 1 neonatal rats. The detrimental effects of alcohol on testicular steroidogenesis can be manifested at the biochemical level in the absence of morphological effects. Acute exposure to alcohol significantly inhibits the catalytic activity of testicular 17-α-hydroxylase in the newborn rat testes. This inhibition was specific since the activity of testicular C17, 20-lyase was not affected. Plasma testosterone levels were reduced to 30% of the control levels in newborn animals receiving al-
cohol. In older animals, i.e. postnatal day 20 and 40 old rats, plasma testosterone levels were reduced, but not significantly. Testicular enzyme activity was not significantly reduced following alcohol treatment in these same older animals. These results suggest that newborn rat tests is especially sensitive to the effects of alcohol (Kelce et al 1990).

The SDN - POA (sexually dimorphic nucleus of the preoptic area of hypothalamus volume of alcohol - exposed males was significantly reduced compared to the pair fed and normal males and became indistinguishable from the SDN-POA volumes of the pair fed and normal females. Alcohol treated females also had a markedly reduced SDN-POA volume compared to the pair fed and normal females. This indicates that the SDN-POA of prepubertal rats of both sexes is sensitive to the effects of in utero alcohol exposure. Plasma testosterone, progesterone and estradiol titers, which were measured in fetuses on gestation day 22, were differentially affected by maternal alcohol consumption, the alterations by themselves cannot adequately explain the effects of prenatal alcohol exposure on the developing SDN-POA (McGivern et al 1984, Barron et al 1988, Ahmed et al 1991).
Central Neurotransmitters:

The main neurotransmitters studied so far are the catecholamines and serotonin. Information is also available regarding glutamate, GABA glycine, Acetylcholine and histamine.

Catecholamines: Low dose of alcohol given to pregnant animals, either in the drinking water or via gastric intubation (Elis et al 1976, Krsiak et al 1977) had little effect on the levels of brain tyrosine (Mena et al 1982) or brain dopamine and norepinephrine (Krsiak et al 1977, Elis et al 1976) in the offspring. High doses of alcohol to pregnant dams generally produced at least transient alterations in brain tyrosine (Detering et al 1980c, Griezerstein et al 1983) and decreased brain dopamine and norepinephrine in the offspring (Rawat, 1977, Detering et al 1980a, 1980c; Shoemaker et al 1983). Although the latter studies reported altered steady-state levels of dopamine and norepinephrine in the brains of the offspring of alcohol-treated animals, other studies did not find catecholamine changes in whole-brain analyses (Jungkuntz-Burgett et al 1990).

Hypothalamic dopamine levels were found to decrease in 21 day old offspring (Detering et al 1980a). The levels were found to be
decreased, in a 19 day old offspring although it was not significant (Rathbun et al 1985). NE was also decreased in the hypothalamus of 3 week old rat offspring. This deficit of NE was seen to persist until 26 weeks after last exposure of the offspring to alcohol (Detering et al 1981). The most plausible explanation for this could be that maternal alcohol consumption results in transiently altered development of noradrenergic synapsis (Thadani et al 1977a; Slotkin et al 1980) and a decreased number of dopamine receptors (Lucchi et al 1983) in the offspring of alcohol treated rats. Fetal alcohol response alters the response of noradrenergic and dopaminergic neurons in the HPOA (Jungkuntz - Burgett et al 1990).

**Serotonin**: Using different methods it has been found that there is very little significance of alcohol on brain tryptophan (Elis et al 1978, Boggan et al 1979, Mena et al 1982). The only significance in these studies include a transient decrease in K+ stimulated release of serotonin from synaptosomes from fetal and young mice (upto 2 weeks) (Boggan et al 1979) and increased tryptophan and serotonin in 4 day old offspring of alcohol water treated rats (Mena et al 1982).
Amino acid neurotransmitters, Acetylcholine, Histamine

Glutamate was found to be decreased in brains of fetal rats when 4g/Kg/d of alcohol was given by intubation (Griezerstein et al 1983). Conflicting reports were obtained when pairfed, liquid diet was used. An increase was seen in brain of fetal, 5 and 10 day old rats (Rawat1977). GABA was seen to increase in the brains of fetal, 5 and 10 days old rats (Rawat 1977). GABA turn over rates were reduced in olfactory tubercles and hypothalamus (Ledig et al 1993) and glycine was decreased in brains of fetal rats (Griezerstein et al 1983). Among the other neurotransmitters, acetylcholine was seen to be decreased in brains of fetal rats (Rawat 1977). Histamine increased in rats of age G1-G10 (Rawat 1980).

Cyclic Nucleotides:

Cyclic nucleotides are of interest in the CNS because they appear to be involved with several aspects of neuronal function, including changes in membrane permeability, regulation of enzymes involved in the synthesis of neurotransmitters, modulation of cell growth and differentiation. Thus the effect of maternal alcohol consumption is of interest. In general, it appears that the activity of adenylate cyclase may be altered (increased in the cerebellum, decreased in the deincephalon).
in some but not all brain regions from fetal and developing rat offspring of dams that consumed alcohol in their drinking water during gestation and lactation (Mena et al. 1982, Salinas et al. 1983). However, since undernutrition could also cause the same effect and because alcohol during lactation causes inhibition of oxytocin, these results are difficult to interpret. Diminished cGMP response to a cholinergic stimulus was seen in the hippocampus of rats whose mothers received alcohol during gestation (West et al. 1981). This is significant because neuronatomical studies have found abnormalities in the offspring of alcohol treated rats (Walker et al. 1980, Barnes et al. 1981, West et al. 1981). Thus fetal alcohol effects are changes in pathological, neutral or beneficial, transient or long term, in normal CNS development that results from alcohol exposure.

There is evidence that differences in genetic sensitivity to alcohol may be a key to alcohol induced fetal damage in cases where some children whose mothers drank heavily throughout their pregnancies appeared unaffected while others are severely damaged (Chernoff et al. 1977, Webster et al. 1980, Gilliam et al. 1985). Identifying the mechanism by which a neuroteratogen alters brain structure function and ultimately behaviour is made difficult by the interrelationships among the
neurochemical, hormonal and other systems involved in the development and maintenance of the brain (Swaab et al. 1984). With alcohol, these difficulties are compounded by the diversity of regulatory and functional systems altered by the acute and chronic administration of alcohol in the adult, by the less well understood changes alcohol causes in these systems in the fetus, by the non-specific nature of ethanol's membrane actions believed to be responsible for its acute contributions of alcohol metabolites to the effects of alcohol, and by complication that may accompany gestational alcohol exposure and by possible differences in alcohol sensitivity.

Acetaldehyde is present to some extent, in human amniotic fluid and both alcohol (Dow et al. 1985) and acetaldehyde (O'shea et al. 1979, Campbell et al. 1983, Beck et al. 1984) can produce growth retardation and birth defects. Reduced caloric intake per se does not produce the deficits in body and brain weight (Weinberg et al. 1984).

Alcohol acts by perturbing the structure of discrete hydrophobic micro environments within brain cell plasma membranes (Chin et al. 1981, Franks et al. 1981, Ingram et al. 1982). The resultant perturbations are thought to lead to conformational changes in membrane bound proteins, altering their function and causing depression of cellular function,
intoxication and anesthesia. Similarities in the patterns of behavioral and neural pathologies caused by alcohol in the adult and fetus suggest that the two effects of alcohol may be related. The ability of prostaglandin synthesis inhibitors both to antagonize some aspects of acute intoxication (George et al 1979) and reduce the effects of fetal alcohol exposure (Randall et al 1983, Randall et al 1984) further support this contention. It may be possible to gain insight into the mechanism of alcohol's teratogenic effects and the location of sensitive brain regions and cellular populations by considering effects and action of alcohol on the adult brain.

Alcohol exposure produces death of existing Purkinje cells in the cerebellum in both neonatal and adult rats (Phillips et al 1982 a ). Withdrawal following chronic alcohol exposure was a critical factor in precipitating cell death (Phillips et al 1984). While the length of alcohol exposure varies considerably with age, the cerebellum seems susceptible to alcohol damage at any age. The results of such comparisons suggest that the areas in the brain sensitive to alcohol induced damage may remain constant from fetal development through adulthood. Pretreatment of ganglioside reduced the neurobehavioural effects (Hungund et al 1993).
Alcohol-induced changes in factors that alter calcium ion metabolism and prostaglandin function, offer two possible interrelated mechanisms that stand out at this time because the evidence supporting them comes from several directions and may be able to explain some of the regional and cell population differences in fetal alcohol sensitivity. Changes in phosphatidylinositol or other Ca\(^{2+}\) handling process could have detrimental effects on neurons that are in the process of neurite growth and synaptic contact development.

Pennington and colleagues (1983) suggested that the mechanism responsible for the characteristic growth suppression observed in humans and animals exposed to large amounts of alcohol during development involved in the suppression of cell development by cAMP. Their hypothesis is that alcohol exposure reduced prostaglandin dehydrogenase activity, producing elevated prostaglandin levels. The increased accumulation of prostaglandin stimulates cAMP which in turn suppresses the rate of cell division, possibly via changes in protein kinases. This prostaglandin hypothesis is supported by Randall et al (1984) who reported that aspirin, which inhibits prostaglandin synthesis antagonised the effects of fetal alcohol exposure by reducing prenatal mortality to control levels and reducing birth defects by approximately 50%.
Maternal alcohol intake would result in intermittent, transient episodes of ischemia and hypoxia for the fetal brain, eventually diminishing the capacity of nerve cells to regulate intracellular calcium. Elevated intracellular calcium becomes the toxic event that leads to changes in glial processes, extracellular K$^+$ ion concentration and cellular excitability, an increased synthesis of arachidonic acid, prostaglandins, leukotrienes and free radicals (Michalis et al 1985). Alcohol induced alterations in the absorption, transport, storage, metabolic activation and exertion of vitamins, trace elements and ions may produce a shortage of these substances even when they are adequately represented in the diet.

Alcohol induced perinatal corticosterone changes are particularly intriguing because corticosterone can directly affect the hippocampus in a number of ways (McEven et al 1985). Treatment of mice with ADH inhibitor and subsequent treatment with alcohol on day 7 increased the prenatal mortality rate and produced external and skeletal malformation in the offspring (Ukita et al 1993). In women who drink heavily in early pregnancy there is a risk of impaired fetal growth (Verkirk et al 1993). In utero ethanol exposure decreases tissue weight in whole brain (Prasad, 1993).
**Gamma Glutamyl Transpeptidase (γ-GT)**

γ-GT is a membrane bound enzyme and is the key enzyme involved in the degradation of glutathione (l-r-glutamyl-L-cysternyl-glycine) (GSH) which is found in all living cells. Glutathione is involved in the maintenance of SH groups of proteins and other molecules, destruction of hydrogen peroxide, other peroxides and free radicals, acts as a catalyst for disulphide exchange reactions and as a coenzyme like glyoxylase. It is involved in the detoxification of foreign compounds and translocation of amino acids across cell membranes (Meister et al 1976). γ-GT is abundant in the kidney, jejunal villi, choroid plexus, salivary glands, bile duct, seminal vesicles, epididymis and ciliary body (Albert et al 1962, Albert et al 1964, Ross et al 1973). γ-GT is said to be involved in the transformation of amino acids across cell membrane where γ-GT acts as a carrier (Meister 1973). The substrates for the enzyme are glutathione (GSH), oxidised glutathione (GSSG), S-substituted glutathione and other γ-glutamyl compounds. An increase in hypothalamic glutathione level was reported in pubertal rats (Pasha and Vijayan 1989). A number of peptides exert synergistic action on the anterior pituitary (Pasha and Vijayan, 1990).
**Lactate Dehydrogenase (LDH)**

Five tetrameric forms of lactate dehydrogenase isozymes are found in mammals. They are LDH - 1 (B4), LDH - 2 (A1,B3), LDH - 3 (A2,B2), LDH - 4 (A3,B1) and LDH - 5 (A4) which are found in different proportions in various tissue. LDH - C4 a tetrameric isozyme is present in mature testes. During testicular development, the LDH-A, -B- and -C- genes are expressed differentially. LDH - A subunits decrease between 2 and 10 days of age while LDH - B subunits increase during this interval. LDH - C4 activity is detected only after 16 days (post partum) (Goldberg et al 1967, Wieben 1981). LDH - C4 is found in spermatogenic cells and has been detected immunohistochemically in spermatocytes (Hintz et al 1977). LDH is localized primarily in the cytosol. A small proportion is localized in mitochondria and on the cell surface of spermatoza (Alvarez et al 1984). The enzyme plays an important role as a shuttle system for NADH between cytoplasm and mitochondria of sperm and may play a role in energy generation and motility of spermatoza (Vandop et al 1977, Blanco 1980).

Mouse oocytes have unusually high levels of LDH activity attributable to LDH - B4. LDH synthesis which represents as much as 18% of total protein synthesis during oocyte growth, decreases during meiotic maturation of oocytes and ferti-
lized eggs (Mangia et al 1975, 1976, Cascio et al 1982). Uterus also has high levels of LDH which is involved in carbohydrate metabolism (Sakhiya et al 1982).
Scope of the Present Study:

From the above review, it can be assumed that alcohol acts at all levels and its effects especially on the reproductive system affects the next generation also. Studies on the hypothalamic γ-glutammyl transpeptidase, an important enzyme involved in the degradation of glutathione is found in all living cells and whose action on hypothalamic and pituitary hormone release is a keen area of current research.

Lactate dehydrogenase (LDH) is another enzyme found in most of the tissues and a specific isomer found in testes is influenced by various agents. Uterus which forms an important part in fetal development also has an isomer of LDH. Hence a study of LDH in male and female could help answer questions on the role of alcohol on the reproductive system.

Thyroid hormones are major hormones involved in growth development and metabolism and the study on the effect of alcohol on thyroid hormones has also been a major area of interest of the parent investigation.