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EFFECTS OF MATERNAL ETHANOL INGESTION ON CEREBRAL NEUROTRANSMITTERS AND CYCLIC-AMP IN THE RAT OFFSPRING

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Abstract—1. To study the effects of maternal alcohol ingestion on brain parameters in offspring, rats were given ethanol for drinking (25% w/v) from the time of mating until sacrifice. Controls drank tap water.

2. Alcohol ingestion reduced daily food and liquid consumption but total caloric intake was only slightly diminished.

3. Maternal body weight increased and offspring body weight, size and brain weight were reduced in the animals receiving alcohol.

4. Brain concentrations of tryptophan, tyrosine and GABA were augmented in ethanol treated mothers at 1 day post-partum.

5. Comparison of brain parameters in offspring of alcoholic mothers with those of controls showed that tryptophan and 5HT concentrations were augmented in 4 day old neonates, NA was increased in 21 day fetuses and 1 day old neonates, and adenylate cyclase activity was also greater in the brains of 21 day fetuses and the cerebellums of 4 day old neonates.

6. Neither phosphodiesterase nor cyclic-AMP concentrations differed in offspring of alcoholic and control mothers.

7. Data showed alterations in brain NA and 5HT systems in the offspring of alcoholic mothers.

INTRODUCTION

Chronic ethanol ingestion during pregnancy in humans produces a syndrome of retarded growth and abnormalities in fetal morphology known as the "fetal alcohol syndrome" (FAS) (Jones *et al.*, 1973). Adverse effects of ethanol on the developing central nervous system have been reported (Rossett, 1979) as reduction of cerebellar tissue, delayed brain myelinization, decreased rate of [^{14}C]leucine incorporation into cerebral ribosomes (Rawat, 1975a; Wunderlich *et al.*, 1979) and changes in the specific activity of several catecholamine enzymes (Branchey & Friedhoff, 1973; Detering *et al.*, 1980). Ethanol is also known to affect monoamine uptake into synaptosomes from fetal and neonatal brains (Thadani *et al.*, 1977) as well as emotional and motor behavior in offspring (Vincent, 1958). Several aspects of the neurochemical mechanisms of FAS are not yet understood. Although ethanol seems to cross the placenta freely (Vorherr, 1974), the rate of ethanol metabolism in the fetus is slower than in the mother (Rossett, 1979) and most of the negative effects of maternal alcohol ingestion in offspring are probably secondary to the metabolic disturbances in the mother.

Several experimental models for FAS have been used (Salaspuro & Lieber, 1980). In the present study, the pregnant rat receiving alcohol in the drinking water was used to determine cerebral neurotransmitter parameters in the offspring. This model is similar to the human situation in that voluntary reduction in food intake occurs which is partially compensated by calories in the ingested alcohol.

Abbreviations: NA = noradrenaline; DA = dopamine; 5HT = 5-hydroxytriptamine; 5-HiAA = 5-hydroxyindole acetic acid.

MATERIALS AND METHODS

Animals

Female Sprague–Dawley rats from our own colony were kept under automatically controlled temperature (25 ± 1°C) and 12 hr light–dark cycles (changing at 9.00 a.m. and 9.00 p.m.). Free access to purina chow diet and drinking water was allowed. After being mated (as shown by the presence of spermatozoids in vagina smears), the rats were placed two in each cage. Those allowed to deliver were housed just after partum in individual cages with their respective pups. Control rats were given tap drinking water *ad libitum* while ethanol treated animals received 15% (w/v) ethanol as drinking solution during the first three days of pregnancy and 25% (w/v) ethanol from the fourth day until sacrifice. Daily food and liquid intake and body weight were measured throughout the experiment between 10.00 and 12.00 a.m. All animals were killed between 10.00 and 12.00 a.m. by decapitation and the fetuses of those sacrificed before parturition were rapidly removed and decapitated. When used for monoamine and amino acid determination, the brains were immediately placed in dry ice and after weighing, they were kept at −80°C until processing. When used for enzyme activity, protein, RNA and DNA evaluations, brains were immediately dissected and kept at −80°C until processing. For cyclic-AMP and cyclic-GMP evaluations, offspring were introduced *in toto* into liquid N_2 and the frozen complete brains were removed and weighed for processing. Blood samples were collected from the wounded necks into heparinized containers and after immediate centrifugation

at 1000 rpm in the cold, plasma was kept at −30°C until processing.

Determinations

Ethanol was tested in plasma by using the method of Bernt & Gutmann (1974).

Monoamines and amino acids. For these determinations, 6 brains from 21 day old fetuses, 5 from 1 day old neonates and 4 from 4 day old neonates were pooled, while the mothers' brains were analyzed individually. For the extraction of monoamines and 5-HiAA, samples were homogenized in 10 ml of ice-cold 0.4 N perchloric acid containing 0.2 ml of 10% EDTA and 0.1 ml of 5% $Na_2S_2O_5$. After centrifugation, supernatants were passed through a strongly acidic cation exchange column (Dowex 50 W-X4) according to Atack & Magnusson (1970). Fluorimetric analyses were made for NA (Bertler *et al.*, 1958); DA (Atack, 1973); and 5HT and 5-HiAA (Atack & Lindquist, 1973). The recovery of the columns was always between 70 and 100% and uncorrected values were used. Another aliquot of the cerebral supernatnant homogenate was lyophilized and resuspended in a 0.15 M lithium citrate buffer pH 2.22 for the analysis of trytophane, tyrosine and γ-aminobutyric acid with a Beckman 121 MB amino acid autoanalyzer (Martín del Rio & Latorre Caballero, 1980).

Enzyme activities. Adenylate cyclase activity was measured in tissue homogenates made in 40 mM Tris-HCl containing 5 mM $MgCl_2$, pH 7.6, with a final protein concentration of 2 mg/ml. A slightly modified version of the method of Farber & Lolley (1976) was used. Briefly, 50–100 μg of sample protein was incubated for 10 min at 30°C in the presence of 40 mM Tris-HCl buffer (pH 7.6), 5 mM $MgCl_2$, 15 mM phosphocreatine, 7 units of creatine kinase, 1 mM cyclic AMP containing 10,000 cpm of (8-^{3}H)-cyclic AMP, 0.08 mM isobutylmethylxanthine and 1 mM ATP containing 1×10^6 cpm of (α-^{32}P)-ATP, in a final volume of 200 μl. The reaction was stopped by adding 50 μl of 200 mM EDTA-disodium salt. The cyclic-^{32}P-AMP formed in the reaction was isolated in neutral alumina columns, eluated with 3.5 ml of 40 mM Tris-HCl buffer (pH 7.6) and its radioactivity was counted. Cyclic AMP phosphodiesterase activity was assayed (Lolley & Farber, 1975) as a coupled reaction with 5'-nucleosidase (0.6 units) by using substrate concentrations 500 or 5 μM when assaying the high or low k_m enzyme respectively. The product was isolated by using AG 1 × 2 slurry resin and the supernatant was counted for radioactivity.

Cyclic nucleotides. Samples were extracted with 0.1 N HCl (Farber & Lolley, 1981) using high specific activity ^{3}H-cyclic AMP as internal marker. Cyclic AMP was measured by the binding protein method of Gilman (1970).

Other determinations. RNA was measured according to the method of Schmidt & Thannhauser (1945) as modified by Munro & Fleck (1967). DNA was extracted in alkaline digests with hot acid and assayed by the method of Wedd & Levy (1955). Proteins were measured (Lowry *et al.*, 1951) by using bovine serum albumin standard.

Statistical evaluation of the data was done by the Student's *t*-test.

RESULTS

Fluid and food consumption

As seen in Table 1, the mean daily consumption of food and drinking fluid through the pregnancy period was significantly lower in rats treated with ethanol than in their controls. In the ethanol group, 29% of the daily total caloric intake corresponded to ethanol but due to reduced food consumption, the total caloric intake was slightly but significantly lower than in control rats (Table 1).

Body weight and size and brain components

Body weight increase throughout pregnancy in the ethanol treated mothers was smaller than in their controls (Fig. 1). This difference became statistically significant from the fourth day of treatment. Maternal brain wet weight did not differ between alcoholics and controls (data not shown) but body weight and size and brain wet weight were significantly reduced in the offspring of alcoholic mothers (Table 2). The reduced brain weight in offspring of alcoholic mothers motivated determination of some index of cellularity. As shown in Table 2, the concentration of protein, DNA and RNA in brain increased progressively from the 21 day fetus to the 4 day old neonate. Neither of these parameters differed in the brains of offspring of alcoholic mothers and controls at any of the ages studied.

Blood ethanol concentration

Great individual variation was found in the plasma levels of ethanol in both mothers (mean values in all the alcohol treated mothers were 9.42 ± 2.46 mM) and their offspring (0.37 ± 0.20 mM) and these values were always higher in the mothers than in their respective offspring ($P < 0.05$).

Brain amino acids

As shown in Table 3, when compared with the controls, brain concentration of tryptophan, tyrosine and GABA were significantly augmented in the alcoholic mothers at day 1 post-partum with no differences at day 21 of gestation or day 4 post-partum. In 21 day fetuses and neonates of 1 and 4 days of age, both tyrosine and tryptophan concentrations in brain were significantly greater than in their respective mothers while GABA concentrations were lower Maternal alcohol ingestion produced an increase in brain tryptophan concentration in the 4 day old neonate and no

Table 1. Effect of chronic ethanol treatment (25% w/v) on fluid and food consumption in pregnant rats

Group	Food intake g/24 hr	Fluid intake ml/24 hr	Total Kcal/ 24 hr
Control $N = 18$	23.01 ± 0.60	30.47 ± 0.54	67.69 ± 1.77
Ethanol $N = 18$	14.68 ± 0.46	13.67 ± 0.40	61.92 ± 0.94
P	<0.001	<0.001	<0.01

Results are means ± SEM. They correspond to the mean values of all treatment periods for each animal.

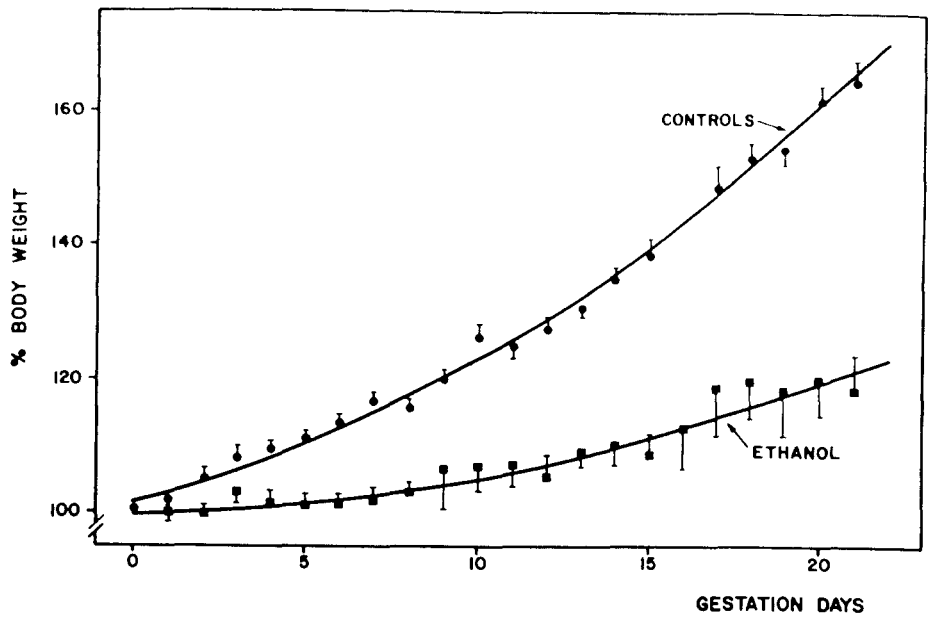

Fig. 1. Body weight increase throughout pregnancy in rats receiving 25% (w/v) ethanol as drinking water as compared with controls receiving tapwater. Values are expressed as percentage of body weight at day 0 of pregnancy. Absolute values at that day were; 211 ± 7 g for controls (n = 18) and 203 ± 8 g for ethanol treated rats (n = 18). P values of the statistical comparison between both groups were <0.001 from the fourth day of treatment.

change in that of the 21 day fetus or 1 day old neonate. Neither tyrosine nor GABA levels in any groups of offspring of alcoholic mothers studied differed from those of control mothers (Table 3).

Brain monoamines and 5HiAA

In mother brains, no significant differences were observed in NA, DA, 5HT or 5HiAA concentrations between ethanol treated rats and their controls (Table 4). From the 21 day fetuses to the 4 day neonates there was a progressive increase in the brain concentrations of NA and DA (Table 5). Both 5HT and 5HiAA brain concentrations rose from the 21 day fetuses to the 1 day neonates but did not increase further in the 4 day neonates. In all the offspring group studied, the brain concentrations of monoamines and 5HiAA were lower than in the respective mothers (Table 5 vs Table 4). Alcohol ingestion in the mothers produced increased NA brain concentration in the 21 day fetuses and 1 day old neonates, while this difference disappeared in 4 day old neonates of alcoholic mothers as compared with controls (Table 5). In these neonates however, the 5HT concentration was enhanced (Table 5), an effect possibly related to the augmented tryptophan concentration also observed in their brains (Table 3).

Brain cyclic nucleotide metabolism in offspring

In Table 6 the results are summarized of cyclic nucleotide metabolism parameters in the offspring of alcoholic and control mothers. Adenylate cyclase activity in the brains of 21 day fetuses was enhanced, in comparison with controls, when mothers were treated with ethanol. This enzyme activity increased rapidly in both groups after birth but there were no differences in the brains of 1 day old or in the forebrains of 4 day old newborns of alcoholic or control mothers. In the 4 day old newborns, adenylate cyclase activity was also measured in the cerebellum (Table 6) and it was significantly higher in the ethanol treated animals than in their controls. Cyclic AMP phosphodiesterase activity of both high and low K_m changed during the perinatal life as already reported (Salinas *et al.*, 1981) but the values found in the brain of offspring of alcoholic mothers did not differ from those of controls. Changes in cyclic AMP concentrations in brain during perinatal life (Table 6) were similar to those previously reported. As shown in Table 6, alcohol ingestion in mothers did not significantly affect these parameters in the brain of offspring. Adenylate cyclase activity was also measured in the brains of alcoholic and control mothers, but no differences were noted between the values of the two groups at 21 days of gestation or 1 or 4 days post-partum (data not shown).

DISCUSSION

Although daily total caloric intake is only slightly decreased by ethanol ingestion in the pregnant rat, it causes a great reduction in maternal body weight gain and a clear retardation in growth indices and brain weight in the offspring. These findings are in agreement with those reported by investigators using different experimental models for maternal alcohol consumption during pregnancy in the rat (Abel & Greizerstein, 1979; Detering *et al.*, 1979, 1980; Rawat, 1975a). Despite a tendency to reduce their whole brain content of protein, DNA and RNA, offspring of alcoholic mothers had parameters not significantly

Table 2. Effect of chronic maternal ethanol treatment (25% w/v) in the rat on growth parameters and brain components in offspring

	Body weight (g)	Body size (cm)	Brain weight (mg)	Protein mg/g	DNA mg/g	RNA mg/g
21 day fetus						
Control	4.19 ± 0.12 (50)	4.49 ± 0.04 (50)	206 ± 3 (52)	61.0 ± 2.2 (8)	3.88 ± 0.31 (5)	3.44 ± 0.1 (5)
Ethanol	3.40 ± 0.13 (66)	3.90 ± 0.08 (69)	179 ± 4 (46)	62.4 ± 2.1 (8)	5.26 ± 0.41 (5)	3.74 ± 0.13 (5)
P	<0.001	<0.001	<0.001	NS	NS	NS
1 day neonate						
Control	6.90 ± 0.11 (51)	5.30 ± 0.05 (52)	287 ± 5 (42)	74.7 ± 3.6 (8)	—	2.98 ± 0.1 (5)
Ethanol	5.51 ± 0.13 (41)	4.91 ± 0.05 (39)	268 ± 4 (40)	63.0 ± 5.1 (8)	3.17 ± 0.13 (5)	2.96 ± 0.04 (5)
P	<0.001	<0.001	<0.01	NS	—	NS
4 day neonate						
Control	9.92 ± 0.13 (64)	6.04 ± 0.04 (64)	424 ± 2 (40)	63.6 ± 2.8 (8)	2.63 ± 0.18 (5)	2.93 ± 0.04 (5)
Ethanol	7.40 ± 0.17 (45)	5.65 ± 0.04 (51)	406 ± 6 (40)	61.8 ± 2.8 (8)	2.68 ± 0.16 (5)	2.93 ± 0.09 (5)
P	<0.001	<0.001	<0.001	NS	NS	NS

Results are means ± SEM (Number of animals in parenthesis).

Table 3. Effect of chronic maternal ethanol treatment (25% w/v) in the rat on brain amino acid concentration in both mothers and offspring

	Mothers			Offspring		
	Tyrosine	Tryptophan	GABA	Tyrosine	Tryptophan	GABA
21 day pregnancy						
Control	8.06 ± 0.93	3.92 ± 0.57	143.4 ± 13	27.15 ± 2.4***	13.31 ± 1.65***	77.2 ± 2.7***
Ethanol	9.11 ± 0.31	4.65 ± 0.32	144.0 ± 7.9	30.79 ± 2.35***	13.59 ± 0.77***	80.5 ± 1.4***
P	NS	NS	NS	NS	NS	NS
1 day post-partum						
Control	6.80 ± 1.11	3.28 ± 0.22	117.1 ± 9.6	16.51 ± 1.44***	10.24 ± 1.87**	78.5 ± 2.9**
Ethanol	13.37 ± 2.06	4.95 ± 0.55	163.9 ± 7.5	12.80 ± 1.92	11.76 ± 2.08*	74.5 ± 4.5***
P	<0.05	<0.05	<0.01	NS	NS	NS
4 day post-partum						
Control	11.25 ± 0.19	3.70 ± 0.11	136.5 ± 4.2	26.99 ± 3.12**	3.81 ± 0.50	91.0 ± 2.6***
Ethanol	10.87 ± 0.40	3.96 ± 0.13	148.3 ± 6.7	25.13 ± 2.63***	7.93 ± 1.36*	98.8 ± 5.2***
P	NS	NS	NS	NS	<0.05	NS

Values are expressed as μg/g of wet weight. Results are means ± SEM of 6–7 determinations per group. Statistical comparison between control and ethanol groups is shown by the *P* values and that between offspring and their respective mothers by asterisks: * = $P < 0.05$; ** = $P < 0.01$; *** = $P < 0.001$.

Table 4. Effect of chronic ethanol treatment on brain levels of monoamines and 5HiAA in pregnant and lactating rats

	NA(ng g)	DA(ng g)	5HT(ng g)	5HiAA(ng g)
1 day pregnancy				
Control	486 ± 23	505 ± 76	242 ± 44	270 ± 58
Ethanol	504 ± 13	525 ± 75	236 ± 45	289 ± 32
1 day post-partum				
Control	515 ± 24	619 ± 54	277 ± 27	235 ± 34
Ethanol	559 ± 13	770 ± 84	281 ± 28	276 ± 49
4 day post-partum				
Control	361 ± 31	695 ± 40	393 ± 10	256 ± 41
Ethanol	341 ± 18	803 ± 39	383 ± 19	295 ± 29

Values are expressed on ng/g of wet weight. Results are means ± SEM of 6 determinations. Statistical comparison between control and ethanol groups gave P values >0.05 (not significant).

Table 5. Effect of chronic maternal ethanol treatment on monoamine and 5HiAA levels in fetal and neonatal rat brain

	NA(ng/g)	DA(ng/g)	5HT(ng/g)	5HiAA(ng/g)
21 day fetus				
Control	66.4 ± 7***	105.1 ± 15.9***	116.6 ± 3.7**	86.5 ± 0.4**
Ethanol	85.5 ± 2.6***	104.4 ± 28.4***	113.1 ± 4.6*	109.8 ± 8.4***
P	<0.001	NS	NS	NS
1 day neonates				
Control	98.2 ± 3.2***	121.7 ± 13.9***	165.8 ± 11.5**	178.9 ± 10.6
Ethanol	110.3 ± 2.3***	129.9 ± 15.4***	160.4 ± 9.8**	182.1 ± 25.5
P	<0.05	NS	NS	NS
4 day neonates				
Control	126.2 ± 4.9***	295.8 ± 24.6***	134.3 ± 5.0***	122.2 ± 14**
Ethanol	128.0 ± 2.8***	260.3 ± 31.9***	169.5 ± 13.0***	164.7 ± 16**
P	NS	NS	<0.05	NS

Values are expressed as ng/g of wet weight. Results are means ± SEM of 6 determinations per group. Statistical comparison between control and ethanol groups is shown by the P values and that between offspring and their respective mothers (data shown in Table 4) by asterisks: * = $P < 0.05$; ** = $P < 0.01$; *** = $P < 0.001$.

Table 6. Effect of chronic maternal ethanol treatment on brain cyclic-AMP related parameters in the offspring

	Adenylate Cyclase pmol/min/mg prot	Cyclic-AMP phosphodiesterase nmol/min/mg prot		Cyclic AMP pmol/mg tissue
		High K_m	Low K_m	
21 day fetus				
Control	169 ± 6	7.43 ± 0.38	0.42 ± 0.02	0.77 ± 0.06
Ethanol	209 ± 11	6.75 ± 034	0.42 ± 0.04	0.79 ± 0.10
P	<0.01	NS	NS	NS
1 day neonate				
Control	265 ± 15	8.05 ± 0.40	0.50 ± 0.03	0.82 ± 0.06
Ethanol	315 ± 36	7.71 ± 0.15	0.47 ± 0.05	0.96 ± 0.09
P	NS	NS	NS	NS
4 day neonate				
Forebrain				
Control	355 ± 19	12.6 ± 0.9	0.48 ± 0.03	1.05 ± 0.13
Ethanol	397 ± 20	11.4 ± 0.5	0.50 ± 0.03	0.82 ± 0.04
P	NS	NS	NS	NS
Cerebellum				
Control	342 ± 24	4.32 ± 0.39	1.22 ± 0.12	
Ethanol	430 ± 24	5.24 ± 0.47	1.40 ± 0.20	
P	<0.05	NS	NS	

Results are means ± SEM of 6–12 determinations.

different from control values, suggesting that brain cellularity development is not greatly impaired in these animals. A certain degree of dehydration may be mainly responsible for the brain weight reduction of offspring of alcoholic mothers, as already demonstrated in adults (Morgan *et al.*, 1957).

Several aspects of the basal comparative changes in neurotransmitter parameters of mothers and their offspring should be considered. Tyrosine and tryptophan brain concentrations in fetuses as compared with their respective mothers at late gestation were augmented as previously reported (Fando *et al.*, 1981) and coincide with the known increase in most amino acids in the fetal as compared with maternal blood (Girard & Marliss, 1975; Palou *et al.*, 1977), probably due to their active transfer through the placenta. Increases in these amino acids were also found in the brains of neonates as compared with their mothers, although circulating amino acids are known to be reduced in newborns (Girard & Marliss, 1975; Girard *et al.*, 1976). There is evidently a specific brain uptake of selected amino acids during the perinatal phase as already described (Braun *et al.*, 1980; Oldendorf, 1971). In spite of this increased brain concentration of neurotransmitter precursor amino acids, the actual amount of neurotransmitters was diminished in offspring during the perinatal phase as compared with their mothers, an affect possibly caused by limited perinatal enzymatic synthesizing activity. At birth, the brain activity of neurotransmitter metabolism enzymes is low and increases progressively with age (Branchey & Friedhoff, 1973; Detering *et al.*, 1980).

Maternal alcohol ingestion affect offspring neurotransmitter metabolism, as shown by the observed changes in their brain concentration. It was previously found that chronic alcohol ingestion in adult rats produced changes in brain monoamine content only in very specific regions (Mena & Herrera, 1980). Thus the appearance of differences in certain biogenic amines and adenylate cyclase activity in whole brain suggest the presence of great alterations in specific brain regions. An increase in brain NA content in the 21 day fetus and 1 day neonate of alcoholic mothers, in the presence of unaltered tyrosine and DA concentrations, would indicate a decreased release of NA more than an enhanced synthesis. We have shown that chronic alcohol ingestion in adult rats does not affect brain catecholamine synthesis whereas NA depletion is retarded after tyrosine-hydroxylase inhibition (Mena & Herrera, 1980). The difference in brain NA concentration between offspring of alcoholic mothers and those of controls disappeared in the 4 day old neonates, but the development of brain structures may not permit detection of the alcohol effects in certain regions. Decreased levels of brain NA and DA have been reported in pups of mothers treated with alcohol (Detering *et al.*, 1980) but differences in the time and size of the administered dose may be responsible for this contradiction with our results. The observed changes in offspring brain NA concentrations produced by ethanol parallel very closely those of adenylate cyclase activity in the presence of unchanged cyclic-AMP phosphodiesterase and cyclic-AMP concentrations. These changes are similar to those reported in adult alcoholic rats in which brain adenylate cyclase activity was augmented (Israel *et al.*, 1972; Kuriyama & Israel, 1973), while cyclic-AMP concentration was unchanged (Redos *et al.*, 1976). It is known, however, that NA activating effects on adenylate cyclase activity are very slight in the immature brain (Schmidt *et al.*, 1970) while it has also been reported that chronic ethanol consumption alters adenylate cyclase hormonal sensitivity (French *et al.*, 1975; Israel *et al.*, 1972). Further studies are required to determine whether maternal alcohol consumption modifies hormonal adenylate cyclase sensitivity in offspring brains.

The increased 5 HT content in the brains of 4 day old rats of alcoholic mothers corresponds to a similar rise in brain tryptophan concentration. We have previously reported indices of increased tryptophan hydroxylase activity in the brains of chronic alcohol-treated adult rats (Mena & Herrera, 1980), and a similar change may occur in the offspring of alcoholic mothers.

The effects of ethanol administration on GABAergic activity are still unclear and the literature contains many contradictions (Hunt & Majchrowicz, 1979; Liljequist & Engel, 1979; Volicer & Klosowicz, 1979). Ethanol drinking throughout pregnancy did not alter brain GABA concentration in offspring and was higher in the maternal brains only at day 1 post-partum. Augmented GABA concentration in the fetal brain has been reported for mothers under alcohol treatment with a liquid diet as compared with sucrose-control rats (Rawat, 1975b). This type of control for neurotransmitter studies has recently been criticized (Badawy *et al.*, 1979) but differences in results may be caused by the different ethanol doses administered.

Changes in brain amino acid levels in alcoholic mothers also deserve some comment. Significantly higher concentrations of the three amino acids studied (tyrosine, tryptopan, and GABA) were only present on the first day post-partum in alcoholic mothers. This effect corresponds more to the reduction in control values, as compared with those before parturition and 4 days post-partum, than to an enhancement in the alcoholics. The reduction in controls could be due to the specific metabolic state of mothers at parturition, involving diminished food intake and increased mammary gland synthesizing activity which produce important decreases in circulating amino acid levels (Bonsnes, 1947). This action may limit the availability of amino acids in the brain until circulating steady state levels are recovered by normal food intake. If this explanation is correct, the decrease in maternal brain amino acids may be caused by ethanol which reduces the activity of amino acid metabolizing enzymes, as reported in other studies (for a review see Shaw & Lieber, 1970), thus protecting the parturating mothers' brains from that temporal reduction in amino acid availability. Further studies are required to confirm this hypothesis.

Observed changes in neurotransmitter metabolism in the offspring of alcoholic rat mothers do not establish their specific relation with the neurological abnormalities described in the FAS (Abel, 1974) but they definitely show the adverse consequences of maternal alcohol consumption on CNS development in offspring.

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