Neurotoxicology and Teratology, Vol. 10, pp. 51–58. © Pergamon Journals Ltd., 1988. Printed in the U.S.A. 0892-0362/88 $3.00 + .00

Dose-Dependent Consequences of Cocaine on Pregnancy Outcome in the Long-Evans Rat

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Received 26 February 1987

CHURCH, M. W., B. A. DINTCHEFF AND P. K. GESSNER. *Dose-dependent consequences of cocaine on pregnancy outcome in the Long-Evans rat.* NEUROTOXICOL TERATOL **10**(1) 51–58, 1988.—The number of obstetric patients abusing cocaine has increased dramatically in recent years. To better understand its effect on pregnancy and to establish the LD50s for maternal and fetal fatalities, the dose-dependent effects of cocaine on pregnancy outcome were investigated in the Long-Evans rat. Pregnant animals were given either saline or 40, 50, 60, 70, 80, or 90 mg/kg cocaine hydrochloride from gestation days 7 to 19 inclusive. An additional group was non-treated and had ad lib access to food and water. Animals were sacrificed on gestation day 20 and the fetuses were examined. Despite treatment during the major periods of organogenesis and brain development, few congenital abnormalities were observed. There were, however, dose-dependent effects on maternal weight gain, maternal food and water consumption, fetal weight, maternal and fetal fatalities, fetal edema, *abruptio placentae* and cephalic hemorrhages. Despite suppression of maternal weight gain, there was preservation of fetal weights at cocaine doses up to and including 80 mg/kg/day, suggesting some protection of fetal growth. In addition to providing information on the gestational effects of cocaine in the rat, the present study provides information useful in guiding the selection of cocaine doses for subsequent behavioral teratology studies.

Abruptio placentae Cocaine Fetal edema Fetal fatality Maternal fatality Pregnancy/pregnant Prenatal Rat Teratogenic/teratology

RECENTLY, there has been an increase in cocaine abuse among obstetric patients and a consequent increase in the number of adversely affected pregnancies. For example, cocaine abuse among obstetric patients has been associated with increased rates of spontaneous abortions and *abruptio placentae* [4, 5, 7]. Prenatal cocaine exposure in humans may also result in an increased incidence of birth defects such as tortuous vessels in the eye [18], congenital heart disease, major and minor craniofacial anomalies [5], and cerebral infarctions [8]. There are also many reports of cocaine-abusing mothers delivering infants of normal weight, gestational age and facial appearance [5, 7, 9, 12, 20]. These infants, however, may later exhibit depression of interactive behavior and poor response to environmental stimuli, raising concerns about developmental delays and permanent central nervous system dysfunction [7,12].

While these human studies have provided useful information about the potentially adverse effects of prenatal cocaine exposure, they have several shortcomings. For example, the human data were collected from small and possibly biased populations. The human data are also clouded by such confounding variables as maternal age and undernutrition, varying concentrations and availability of cocaine, multiple drug use, poor pre- and post-natal care, and the presence of toxic adulterants that are mixed with or used to process cocaine (e.g., amphetamines, procaine, talc, solvents, heroin, phen-

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cyclidine) [6]. Thus, it is uncertain whether cocaine is entirely responsible for these adverse obstetrical outcomes or whether they were due, in whole or in part, to other factors.

Animal studies, on the other hand, enable one to control for many of these confounding variables. Yet, only a handful of animal studies on prenatal cocaine exposure have been conducted and with conflicting results. For example, one group of researchers found that prenatal cocaine exposure was highly teratogenic in CF-1 mice, but had no influence on fetal weight and maternal weight gain [21,22]. In contrast, another group found only minor evidence of cocaine-induced *terata* in Sprague-Dawley rats and Swiss Webster mice, but did observe strong adverse effects on fetal weight, maternal weight gain, resorptions, and fetal edema [14].

In addition to conflicting results, the above animal studies had a number of limitations. First, the maternal and fetal toxic effects were not evaluated over a broad range of cocaine doses. Consequently, it is uncertain how much cocaine is needed to observe an adverse effect on pregnancy outcome and what effects (e.g., resorptions, teratologies, maternal and fetal fatalities) occur at a particular dose. Secondly, cocaine was administered to animals during a rather limited period of gestation. Since cocaine-addicted individuals use the drug daily when availability permits, administration of cocaine daily over a prolonged period during pregnancy might provide a more pertinent model. Thirdly, Mahalik and colleagues [21,22] gave cocaine doses to pregnant rats with dose selection based on the toxic effects observed in non-pregnant animals, while Fantel and MacPhail [14] etherized animals prior to intraperitoneal cocaine injections. These procedures have obvious shortcomings. For example, pregnancy alters the toxicity [17], blood levels [1] and other pharmacokinetic aspects [29] of drugs in rodents. This is particularly true for cocaine [29]. For these reasons, it is recommended that the dose selection for maternal and fetal toxicology studies be based on preliminary findings with pregnant, rather than non-pregnant, animals [1,17]. Further, etherizing animals on a daily basis prior to cocaine administration [14] resulted in an ether-by-cocaine interaction study, rather than a cocaine study *per se*. Various ethers have reproductive toxicity [16], and easily pass the placental barrier [26]. They can also cause hypoxia, neurotoxicity, and liver enzyme induction. Finally, intraperitoneal cocaine injections [14] can conceivably cause maternal and fetal death by peritonitis or direct trauma to the fetuses rather than by cocaine toxicity *per se* [24].

Considering these issues, the present study sought to investigate the dose-dependent effects of cocaine on pregnancy outcome in the rat, administering cocaine subcutaneously throughout most of the gestational period. One goal was to see if the reported effects of cocaine on human pregnancy (e.g., *abruptio placentae*, stillbirths) could be reliably reproduced in an animal model. A second goal was to determine which cocaine dose levels would cause morphological abnormalities in the conceptus and to establish dose-response curves for maternal and fetal fatalities and other indices of gestational toxicity (e.g., impaired maternal and fetal weight gain, impaired maternal food/water consumption). This information does not exist for the Long-Evans rat and should benefit researchers undertaking behavioral teratology studies by indicating which cocaine doses are excessively toxic to this species in terms of pregnancy outcome.

METHOD

Timed-pregnant nulliparous female Long-Evans hooded rats, aged 90 days at the time of mating, were housed in Plexiglas cages (45×23×20 cm) with beddings of wood shavings. Rooms were temperature (22±1°C) and humidity (40–50%) controlled with a timed light cycle of 14 hr of light per day (5:00 a.m. to 7:00 p.m.).

Matching for weights, animals were assigned to an ad lib control group, a saline control group, or one of six cocaine treatment groups. The morning on which a sperm plug was found was designated gestation day 0. Starting on gestation day 7 and continuing to gestation day 19 (inclusive), the rats in the cocaine treatment groups were given subcutaneous injections of 40, 50, 60, 70, 80 or 90 mg/kg cocaine hydrochloride (HCl) dissolved in distilled water (2% solution). Gestation days 7–19 include the major periods for organogenesis and CNS development in the rat [32]. Cocaine doses were selected on the basis of the literature and the desire to choose dose levels that would adequately span the range of fetal and maternal lethality [14, 21, 22, 24]. Saline doses were isovolumetric to the 90 mg/kg cocaine doses. Each daily dose of cocaine or saline was split evenly with the first portion administered between 9–10 a.m. and the second portion administered between 3–4 p.m. To obviate problems associated with skin ulcerations, cocaine solution volumes greater than 0.30 ml were injected into multiple sites. While some skin ulcerations did occur in the higher dose groups (60 mg/kg and above), these were treated with antibiotic ointment and healed rapidly. We avoided injecting the cocaine solution in the nape of the neck since this region seemed unusually sensitive to skin ulcerations.

Daily records were maintained on maternal weights and food/water consumption. Initially, 5 or 6 dams were assigned to each treatment group. Some animals eventually proved not to be pregnant, lowering some group sizes to n=4. A second control group of non-treated, ad lib fed and watered animals (n=11) was also employed for comparison purposes. The pregnant dams were sacrificed by carbon dioxide inhalation on gestation day 20 (term=day 21). The fetuses were removed from the uterine horns, counted and weighed. The uteri were subsequently placed in a solution of ammonium sulfide and saline (10% w/v). The next day, the stained uteri were placed between glass slides and examined for implantation sites. Most data were first analyzed by one-way analyses of variances (ANOVAs). When an ANOVA indicated a significant main effect, simple effects tests (*post hoc* *t*-tests) were then conducted to determine which treatment group(s) differed significantly from the two control groups. Fatality-dose relationships were evaluated by probit and log linear analyses (available on SPSS-X software). Statistical significance was assumed for probability levels of 0.05 or less (non-directional tests).

Frequently, studies on the effects of prenatal drug exposure employ one of several pair-feeding/pair-watering techniques to permit the differentiation of the effects of drug plus undernutrition from the effects of undernutrition alone [1–3]. This is a meritorious procedure for behavioral teratology studies, but not necessarily appropriate for gestational toxicology studies. For example, since maternal lethality was being investigated, pair-feeding a dam to one that was expected to die was impractical. Moreover, limited funding prevented the use of a pair-fed control group for each cocaine group. Pair-feeding the lower dose groups to animals in the highest dose group economizes on animal usage. How-

TABLE 1

DOSE-DEPENDENT EFFECTS OF COCAINE ON PREGNANCY OUTCOME IN THE RAT: MATERNAL DATA

		Treatment Group/Daily Cocaine Dose							
	Variable	Ad Lib	Saline	40 mg/kg	50 mg/kg	60 mg/kg	70 mg/kg	80 mg/kg	90 mg/kg
No. pregnant dams at treatment onset		11	5	6	4	6	5	6	6
No. pregnant dams surviving treatment		11	5	6	4	4	3	3	1
Maternal fatality Rate (%)		0	0	0	0	33	40	50	83
Maternal weight gain (g)	mean	91	86	66*	61*	47*	43*	18*	19*
	SD	11	34	11	4	15	8	11	7
Daily food consumption (g)	mean	20.0	18.6	15.3*	15.9	13.2*	13.6*	11.8*	8.5*
	SD	2.4	1.7	2.0	2.4	1.0	2.7	1.1	4.5
Daily water consumption (ml)	mean	43.1	33.5	38.7	45.4	32.5	32.0	34.8	26.2*
	SD	6.0	3.3	5.3	3.3	3.1	5.7	5.9	9.6

*Significantly less than Ad Lib and Saline control groups' averages, $p=0.05$ level or better.

ever, such a pair-feeding technique adds intentional food restriction as a confounding variable to any observed effect. Consequently, all animals were allowed ad lib access to food and water for this preliminary study.

For the sake of brevity, the various cocaine treatment groups will hereafter be referred to as COC40, COC50, COC60, etc.

RESULTS

Maternal Fatality

A significant, $F(1,4)=24.30$, $p<0.005$, dose-dependent increase in maternal mortality was observed (see Table 1). The threshold for maternal lethality was between 50 and 60 mg/kg/day cocaine HCl. Probit analysis estimated the median lethal dose (LD50) to be 74 mg/kg/day cocaine HCl (95% confidence limits=65–85 mg/kg/day). There was a total of 12 cocaine-induced maternal fatalities among the 23 dams receiving 60 or more mg/kg/day. Ten of the 12 fatalities (83%) occurred late in the treatment period (gestation days 17–19 inclusive; i.e., after 11–13 days of treatment).

Maternal Weight Gain

There was no significant difference in maternal weights prior to treatment. For example, on gestation day 7 (before treatment began), the average (±S.D.) weights for the dams in the Ad Lib, Saline, COC40, COC50, COC60, COC70, COC80 and COC90 groups were 296±23, 285±26, 271±17, 294±27, 272±15, 273±14, 268±18 and 270±22 g, respectively, $F(7,41)=2.022$, $p>0.05$. On gestation day 20, after 13 consecutive days of cocaine treatment, there was a definite dose-dependent difference in maternal weight gain relative to treatment onset (i.e., relative to gestation day 7-weights), $F(7,30)=13.344$, $p<0.001$. The data in Table 1 indicate that there was progressively less maternal weight gain with increasing cocaine dose. The data in Table 1 also indicate that even the lowest cocaine dose (40 mg/kg/day) significantly affected maternal weight gain. One phenomenon that is not entirely reflected by the data in Table 1 is that an extreme and sudden weight loss of 20–30 g was usually observed in any given dam the day prior to a cocaine-induced fatality.

Maternal Food and Water Consumption

There was no significant group difference in daily food consumption prior to treatment. For example, the average (±S.D.) daily food consumption prior to treatment for dams in the Ad Lib, Saline, COC40, COC50, COC60, COC70, COC80 and COC90 groups were 18.4±2.9, 18.0±2.4, 17.5±2.3, 19.8±1.7, 18.4±2.9, 17.5±2.0, 16.0±1.6 and 18.5±1.8 g, respectively, $F(7,41)=1.160$, $p>0.10$. Similarly, prior to treatment, there was no significant group differences in average (±S.D.) daily water consumption: 34.7±4.6, 34.6±4.1, 32.5±6.9, 33.8±4.6, 30.4±5.7, 31.4±6.6, 30.0±4.9 and 31.1±7.9 ml, respectively, $F(7,41)=0.715$, $p>0.10$.

During the treatment period, however, there was a significant group-dependent difference in average daily food consumption, $F(7,41)=16.513$, $p<0.001$. The mean values in Table 1 indicate a more or less progressive decrease in food consumption with increasing cocaine dose. This decrease in food consumption was marginal for the COC40 and COC50 groups. While the decrease in food consumption was greater for higher cocaine dose groups, this trend was not always dramatic (see Table 1). Cocaine administration also had a significant effect on daily water consumption, $F(7,41)=7.227$, $p<0.001$. However, the data in Table 1 indicate that a significant decrease in daily water consumption occurred only for the highest dose group (i.e., COC90).

Conceptus Fatality

The data on fetal outcome are displayed in Table 2. Including the data from all dams, there was no significant group difference in the average number of uterine implant sites, $F(7,41)=0.913$, $p>0.10$ (see Table 2). There was, however, a significant dose-dependent decrease in the number of

TABLE 2

DOSE-DEPENDENT EFFECTS OF COCAINE ON PREGNANCY OUTCOME IN THE RAT: FETAL DATA

Variable		Treatment Group/Daily Cocaine Dose							
		Ad Lib	Saline	40 mg/kg	50 mg/kg	60 mg/kg	70 mg/kg	80 mg/kg	90 mg/kg
No. litters at treatment onset		11	5	6	4	6	5	6	6
No. litters surviving treatment		11	4*	6	4	4	3	2	1
Implants	n	143	53	69	57	79	58	63	77
	mean	13.0	10.6	11.5	14.3	13.2	11.6	10.5	12.8
	SD	2.0	5.6	0.8	3.4	4.0	3.1	3.0	3.4
Fetuses surviving treatment	n	130	49	63	56	56	25	14	12
	mean	11.8	9.8	10.5	14.0	9.3	5.0†	2.3†	2.0†
	SD	2.0	5.7	1.5	3.8	7.6	4.6	4.1	4.9
Conceptus fatality rate (%)		9	8	9	2	29‡	57‡	78‡	84‡
Fetal weights (g)	mean	3.4	3.8	4.0	3.5	3.5	4.0	3.7	2.8†
	SD	0.4	0.3	0.2	0.3	0.3	0.2	0.3	0.0
Abruptio placentae									
No. fetuses		0	0	0	0	15	0	8	1
No. litters		0	0	0	0	1	0	1	1
Fetal edema									
No. fetuses		0	0	0	0	2	3	17	1
No. litters		0	0	0	0	1	1	3	1
Hemorrhage									
No. fetuses		0	0	1	1	5	4	10	2
No. litters		0	0	1	1	2	1	3	2
No. fetuses with birth defects		0	0	1	0	0	0	0	1

*One Saline female had a single implant site, but no fetuses.
†Significantly less than Ad Lib and Saline control groups' averages, $p=0.05$ level or better (post hoc t-tests).
‡Significantly greater than Ad Lib and Saline control values, $p=0.002$ level or better (Chi Square tests).

live fetuses at gestation day 20, $F(7,41)=5.991$, $p<0.001$. *Post hoc* t-tests indicated that significant decreases in the number of live fetuses by gestation day 20 occurred for the COC70 and higher cocaine treatment groups (see Table 2).

To better determine the relationship between cocaine dose and conceptus fatality, the total number of resorptions + fetal deaths in each treatment group was divided by the total number of implant sites to derive an incidence rate of conceptus fatality. These values are displayed in Table 2 and show a definite increase in the incidence rate of conceptus fatalities with increasing cocaine dose, $F(1,4)=118.69$, $p<0.001$. Combining the two sets of control data to allow an estimate of natural mortality (9%), the LD50 for conceptus loss was estimated by probit analysis to be 71 mg/kg/day cocaine HCl (95% confidence limits=68–74 mg/kg/day). This value was not significantly different from the LD50 of 74 mg/kg/day for maternal fatality. This is not surprising because 152 of 177 (86%) of the conceptus losses were associated with maternal fatalities. Of the remaining 25 losses, 11 were fetal deaths occurring in dams surviving cocaine treatment (1 fetal death in a COC60 litter, 10 fetal deaths in a COC80 litter). These losses occurred near the end of the treatment period in well developed fetuses and were associated with *abruptio placentae* and/or fetal edema (i.e., they would have been stillborn). The remaining conceptus losses (n=14) seemed to represent a natural incidence of embryonic resorptions. Chi square tests on the incidence rates of conceptus fatality indicated that a significant increase in conceptus fatality was seen starting with the COC60 group (see Table 2).

Fetal Weight

Using only data from litters that were still alive on gestation day 20 (litter mean value=unit of measure), there was a significant group-dependent decrease in average fetal weights, $F(7,27)=4.180$, $p=0.003$. The *post hoc* tests indicated that only the highest cocaine treatment group (i.e., COC90) had a significant decrease in average fetal weight, but only in comparison with the Saline Control group ($p=0.012$). The average fetal weights of the other cocaine treatment groups were well within the range of normalcy as evidenced by the Saline and Ad Lib control groups' values.

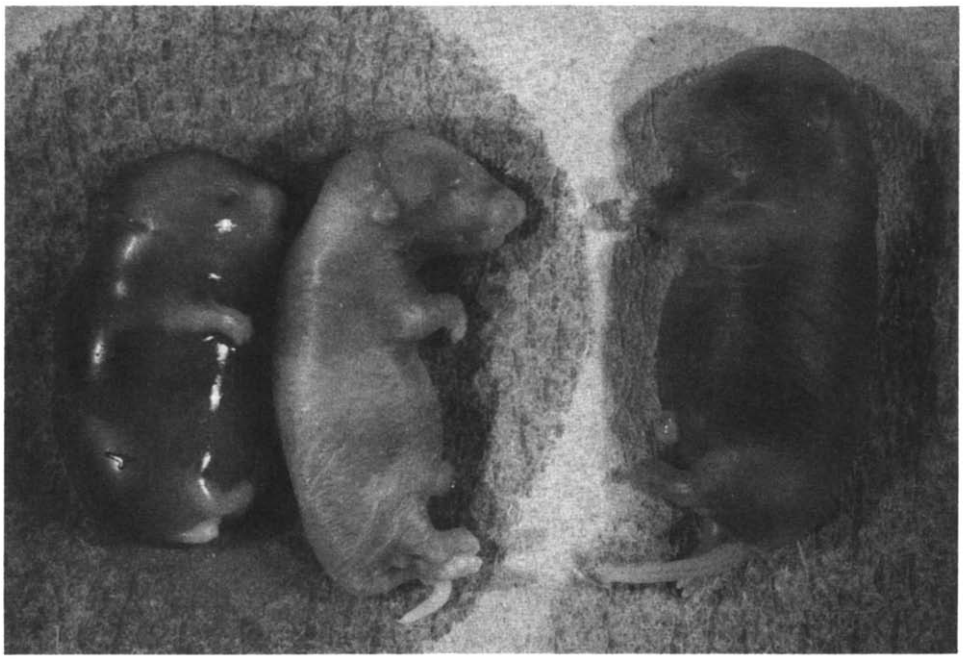

FIG. 1. Left: one edematous and growth retarded rat fetus from the COC90 treatment group on gestation day 20 compared to a non-edematous but growth retarded littermate. Right: one edematous fetus from the COC80 treatment group on gestation day 20.

Because only one COC90 litter was still alive at the time of sacrifice on gestation day 20, the implied trend for fetal weight reduction in this treatment group is tenuous. To further assess the possibility of a reduction in fetal weight in the COC90 group, a second ANOVA was done. This time it included data from the 2 litters (1 each from the COC80 COC90 treatment groups) that came from dams that were alive on the evening of gestation day 19, but were found dead the next morning (gestation day 20). This second ANOVA also showed a significant dose-dependent effect, $F(7,29)=5.840, p<0.001$. With this second analysis, *post hoc* tests found that the fetal weight (mean±S.D.=2.65±0.18 g) for the COC90 group (n=2) was significantly less than both control groups and all other cocaine treatment groups (all p values <0.01 or better). The average fetal weights of these additional COC80 and COC90 litters were 3.53 and 2.52 g, respectively. Consequently, the weights of the fetuses from the litters found dead on gestation day 20 were comparable to those of their live cohorts (see Table 2). These additional analyses strengthen the suggestion that the highest cocaine dose influenced fetal weight, causing a weight reduction of about 25%.

Abruptio Placentae *and Fetal Edema*

Abruptio placentae (as evidenced by vaginal bleeding and retroplacental clots) were confined to the higher cocaine treatment groups (COC60 and above). Placental abruptions did not occur in any of the 130 Ad Lib or 49 Saline control fetuses, but were observed in 4 cocaine litters and involved 24 of the 360 near-term fetuses, $p=0.001$ (Fisher's Exact Test). The incidences of placental abruption for each treatment group are displayed in Table 2. All of the placental abruptions occurred late in the treatment period, between gestation days 16 and 19.

Fetal edema was also confined to the higher cocaine treatment groups (COC60 and above). Fetal edema involved 23 fetuses in 6 litters. Fetal edema did not occur in the control groups ($p<0.001$). The incidences of fetal edema for each treatment group are displayed in Table 2. Figure 1 shows examples of fetal edema.

Hemorrhages and Physical Anomalies

Hemorrhagic areas, easily observed grossly, were seen in 10 cocaine litters and involved a total of 23 fetuses. The majority of these fetuses came from the higher dose groups (COC60 and above). These fetuses comprised a group largely exclusive of those with edema. Hemorrhages did not occur in the Ad Lib or Saline control fetuses ($p<0.001$). The majority of the hemorrhages were in the cephalic region. The incidences of fetuses with hemorrhages for each treatment group are displayed in Table 2. Sixteen of the 23 fetuses with hemorrhages were from pregnancies that terminated prematurely in maternal fatality. Typically, fetuses with hemorrhages (and most of their littermates) exhibited pronounced vasodilation about the head and face. Examples of fetuses with hemorrhages and pronounced vasodilation are shown in Fig. 2.

There were only 2 incidences of physical anomalies in the 360 near-term fetuses from the 33 cocaine litters. One instance of unilateral anophthalmia occurred in a COC40 fetus, and one instance of microcephaly occurred in a COC90 fetus. No birth defects were seen in any of the Ad Lib or Saline control fetuses ($p=0.45$).

DISCUSSION

The present study found that cocaine-induced maternal and fetal mortality developed almost simultaneously in the Long-Evans rat. For example, maternal and fetal LD50's were estimated at 74 and 71 mg/kg/day when cocaine was administered subcutaneously from gestation days 7–19 in-

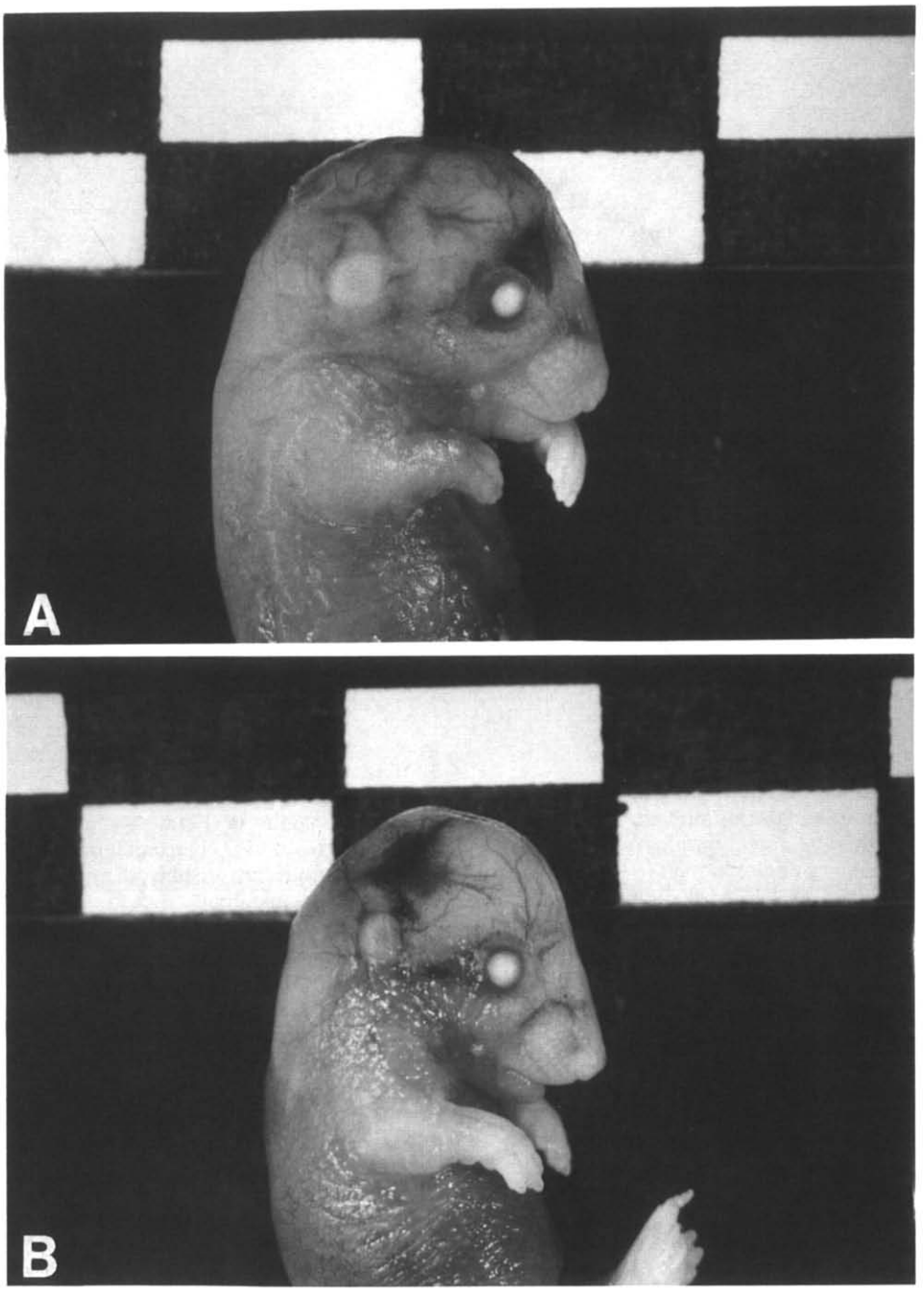

FIG. 2. (A) an example of a cephalic hemorrhage above the eye of a rat fetus from the COC60 treatment group (gestation day 19). (B) an example of a cephalic hemorrhage above the auricle of a rat fetus from the COC70 treatment group (gestation day 18).

clusively with half of the daily dose given in the morning and half in the afternoon. Also, the thresholds for increased maternal and fetal lethality were between 50 and 60 mg/kg/day. One goal of the present study was to provide information for making dose selections in subsequent behavioral teratology studies in the Long-Evans rat. It has been suggested that behavioral teratology studies employ several dose levels with the highest dose causing low rates of malformations and/or deaths *in utero* [34–36]. Based on these criteria, the present findings suggest that a dose between 50–60 mg/kg/day might be suitable for the maximum dose in behavioral teratology studies. Maternal and fetal mortality seemed highly dependent on the number of days of cocaine administration, however. That is, most cocaine-related fetal and maternal deaths occurred late in the treatment period, between 10–13 days of treatment. Had the treatment period been reduced (or increased) by a few days, the maternal and fetal mortality rates might have been greatly altered. It is also very likely that time during gestation is an important factor in cocaine's effect on pregnancy. For example, pregnant rats (and possibly humans) might be most susceptible to cocaine-induced placental abruption late in gestation. Thus,

cocaine dose selection for behavioral teratology studies may depend greatly on both the duration of the treatment period and the gestational age, in addition to one's criteria for acceptable rates of mortalities and physical anomalies.

While the majority of fetal fatalities occurred in association with maternal deaths, there were instances in which pregnant cocaine-treated dams survived to the end of the experiment and were found carrying dead fetuses as evidenced by pronounced fetal edema, *abruptio placentae*, autolysis, and hemorrhages. The condition of these fetuses suggested that death occurred within the last day or two of the treatment period. In other words, the cocaine treatment would have caused some stillbirths. Thus, our findings in the Long-Evans rat agree with the suggestion that cocaine abuse by pregnant women can increase the incidence of stillbirths [7].

Ten of the 12 maternal fatalities occurred towards the end of the treatment period (gestation days 17–19, inclusive) after 11–13 days of cocaine treatment. This is not surprising since repeated cocaine use is associated with increased susceptibility to seizures and death [13,30]. Two common causes of death following repeated cocaine use are (a) brain seizures and (b) respiratory failure secondary to medullary depression [28]. It is of interest that on the day prior to death, most females showed a precipitous weight loss and profound lethargy prior to the morning injection. These events might forewarn fetal and/or maternal death in the human situation as well.

Weight gain, like mortality, provides a measure of the maternal-fetal toxicity relationship. The present study found that cocaine-induced reductions in fetal and maternal weight gain *did not* develop simultaneously. For example, while cocaine doses as low as 40 mg/kg/day adversely affected maternal weight gain, it required a dose of 90 mg/kg/day to significantly influence fetal weight gain. Utilization of stored maternal nutrients is one possible mechanism by which fetal weight gain was protected. Cocaine-induced vasoconstriction at the uterus or placenta, reducing fetal exposure to cocaine [37], might have been another protective mechanism. In this regard, it is of interest that cocaine levels in fetal tissue are significantly less than in maternal tissue [29]. In any event, the fact that normal fetal weights were usually observed is consistent with reports that women who abuse cocaine usually deliver infants who are ostensibly normal by birth weight and gestational age [5, 7, 9, 12, 20].

The anorexic effects of cocaine are well-known [6, 25, 33] and cocaine-induced effects on maternal food consumption were observed in the present study. The cocaine-induced reduction in maternal food consumption developed, more or less, simultaneously with reduced maternal weight gain. That is, significant decreases in daily food consumption (like maternal weight gain) were noted at the lowest dose level of 40 mg/kg/day. Yet, the effect of cocaine on food consumption was not as severe as on maternal weight gain. For example, the COC80 group's maternal weight gain during treatment was about 20% of normal, while daily food consumption was only about 60% of normal. The dose-response for cocaine-induced suppression of food consumption, moreover, was not as severe as that associated with equally lethal alcohol or alcohol-plus-cocaine doses [10,11]. Perhaps the state of pregnancy ameliorated cocaine's anorexic effects or perhaps previous accounts of cocaine-induced anorexia were over-stated. In any event, the fact that cocaine's effects on maternal weight gain were stronger than food consumption suggests that maternal food utilization/excretion were greater than intake. The effect of cocaine on water consumption was even less dramatic than for food consumption. That is, a significant decrease in daily water consumption was only observed for the highest cocaine group (90 mg/kg/day). The fact that cocaine can cause significant dose-dependent effects on maternal weight gain and food/water consumption is of interest because these factors can have important influences on maternal and fetal mortality.

The present study also observed fetal edema, placental abruption, and hemorrhagic areas in cocaine-treated litters, usually at daily doses of 60 mg/kg or more. Our findings of fetal edema, placental abruption, and hemorrhagic areas confirm the reports of others. That is, prior research in the rat has observed incidences of fetal edema in association with prenatal cocaine exposure [14]; and research in humans has observed incidences of placental abruption [4, 5, 7], perinatal cephalic hemorrhages [20], and perinatal cerebral infarctions [8] in association with maternal cocaine abuse. Cocaine use can cause transient hypertension [25,37]. Hypertension, in turn, can cause placental abruption [27], fetal edema [15], prenatal hypoxia and cerebral infarctions in humans [19, 23, 31]. Indeed, a recent study using pregnant ewes found that cocaine produced dose-dependent increases in maternal blood pressure and decreases in uterine blood flow. These responses were accompanied by marked fetal hypoxemia, hypertension, and tachycardia [37]. It is also of interest that chronic cocaine administration is associated with vitamin C deficiency, which increases the susceptibility to hemorrhaging [33].

In terms of cocaine's teratogenic potential, only 2 out of 360 near-term cocaine-exposed fetuses had birth anomalies. One fetus, from the 90 mg/kg/day group, was microcephalic. This may have resulted from restricted intra-uterine space rather than cocaine embryotoxicity. The other birth anomaly, from a fetus in the 40 mg/kg/day group, involved unilateral anophthalmia. Since this fetus was also growth retarded (weighing only 2.4 grams on gestation day 20), it is possible that this fetus had suffered from cocaine toxicity. Interestingly, one research group [21,22] reported anophthalmia as a common finding in CF-1 mice exposed prenatally to cocaine. Considering the relatively low incidence rate of anomalies, however, we must conclude that cocaine has low teratogenic potential in regards to the external appearance of the Long-Evans rat. Similarly, another research group [14] found little evidence of cocaine teratogenicity in Sprague-Dawley rats and Swiss-Webster mice. Cocaine also seems to have little effect on human neonates with regards to external appearance [4, 5, 7, 9, 12, 20].

In summary, cocaine was found to adversely influence maternal weight gain, food consumption and mortality as well as fetal weight and mortality. Cocaine-induced maternal mortality developed almost simultaneously with fetal mortality. Fetal weight reduction did not develop simultaneously with impaired maternal weight gain and food consumption. Cocaine was also associated with placental abruption, fetal edema, fetal hemorrhages and fetal and maternal death in a well-controlled animal experiment—thus lending credence to similar findings in earlier human and animal studies that were clouded by confounding variables (see Introduction). These manifestations of toxicity (or toxicity plus undernutrition) showed dose-dependent relations. That is, under this study's treatment protocol, the thresholds for many of the more serious adverse effects (e.g., mortality, fetal edema, hemorrhages, stillbirths, *abruptio placentae*) occurred at daily

doses of 50–60 mg/kg. Our findings, while chiefly of interest to developmental toxicology research in animal models, are also relevant to the clinical situation because they suggest that maternal cocaine use places the human fetus at risk for low birth weight, edema, placental abruption, infarctions and death.

ACKNOWLEDGEMENTS

We thank K. Schmid for photographic illustrations and K. Callanan for manuscript preparation. This project was supported in part by NIDA grant RO1 DAO5536 awarded to Dr. Church and by BRSG S07 05938-02 awarded by the Biomedical Research Support Grant Program, Division of Research Resources, National Institutes of Health.

REFERENCES

1. Abel, E. L. Procedural considerations in evaluating parental effects of alcohol in animals. *Neurobehav Toxicol* **2:** 167–174, 1980.
2. Abel, E. L. and B. A. Dintcheff. Factors affecting the outcome of maternal alcohol exposure: I. Parity. *Neurobehav Toxicol Teratol* **6:** 373–377, 1984.
3. Abel, E. L. and B. A. Dintcheff. Factors affecting the outcome of maternal alcohol exposure: II. Maternal Age. *Neurobehav Toxicol Teratol* **7:** 263–266, 1985.
4. Acker, D., B. P. Sachs, J. K. Tracey and W. E. Wise. Abruptio placentae associated with cocaine use. *Am J Obstet Gynecol* **146:** 220–221, 1983.
5. Bingol, N., M. Fuchs, V. Diaz, R. K. Stone and D. S. Gromisch. Teratogenicity of cocaine in humans. *J Pediatr* **110:** 93–96, 1987.
6. Byck, R. and C. Van Dyke. What are the effects of cocaine in man? In: *Cocaine: 1977,* NIDA Monograph No. 13, edited by R. C. Petersen and R. C. Stillman. Washington, DC: DHEW, 1977, pp. 97–118.
7. Chasnoff, I. J., W. J. Burns, S. H. Schnoll and K. A. Burns. Cocaine use in pregnancy. *N Engl J Med* **313:** 666–669, 1985.
8. Chasnoff, I. J., M. E. Bussey, R. Sarich and C. M. Stack. Perinatal cerebral infarction and maternal cocaine use. *J Pediatr* **108:** 456–459, 1986.
9. Chasnoff, I., C. Hunt, R. Kletter and D. Kaplan. Increased risk of SIDS and respiratory pattern abnormalities in cocaine-exposed infants. *Pediatr Res* **20:** 425A, 1986.
10. Church, M. W., B. A. Dintcheff and P. K. Gessner. Alcohol, cocaine and pregnancy in the rat: II. Fetal data. *Alcohol: Clin Exp Res* **11:** 196, 1987.
11. Dintcheff, B. A., M. W. Church and P. K. Gessner. Alcohol, cocaine and pregnancy in the rat: I. Maternal data. *Alcohol: Clin Exp Res* **11:** 197, 1987.
12. Dougherty, R. J. "Is cocaine use an alcoholism problem?" Presented at a conference sponsored by the New York State Division of Alcoholism and Alcohol Abuse, White Plains, NY: November 6, 1985.
13. Downs, A. W. and N. B. Eddy. The effect of repeated doses of cocaine in the rat. *J Pharmacol Exp Ther* **46:** 199–200, 1932.
14. Fantel, A. C. and B. J. MacPhail. The teratogenicity of cocaine. *Teratology* **26:** 17–19, 1982.
15. Grabowski, C. T. Altered electrolyte and fluid balance. In: *Handbook of Teratology* Vol II, edited by J. G. Wilson and F. C. Fraser. New York: Plenum Press, 1977, pp. 153–170.
16. Hardin, B. D. Reproductive toxicology of the glycol ethers. *Toxicology* **27:** 91–102, 1983.
17. Hutchings, D. E. Prenatal opioid exposure and the problem of causal inference. In: *Current Research on the Consequences of Maternal Drug Abuse,* NIDA Monograph No. 59, edited by T. M. Pinkert. Washington, DC: DHHS, 1985, pp. 6–19.
18. Isenberg, S. J., A. Spierer and S. H. Inkelis. Ocular signs of cocaine intoxication in neonates. *Am J Opthalamol* **103:** 211–214, 1987.
19. Korones, S. B. and J. Lancaster. *High-Risk Newborn Infants: The Basis for Intensive Nursing Care.* St. Louis: C. V. Mosby Co., 1981.
20. Madden, J. D., T. F. Payne and S. Miller. Maternal cocaine abuse and effect on the newborn. *Pediatrics* **77:** 209–211, 1986.
21. Mahalik, M. P., R. F. Gautieri and D. E. Mann, Jr. Teratogenic potential of cocaine hydrochloride in CF-1 mice. *J Pharm Sci* **69:** 703–706, 1980.
22. Mahalik, M. P., R. F. Gautier and D. E. Mann, Jr. Mechanisms of cocaine-induced teratogenesis. *Res Commun Subst Abuse* **5:** 279–302, 1984.
23. Ment, L. R., C. C. Duncan and R. A. Ehrenkranz. Perinatal cerebral infarctions. *Ann Neurol* **16:** 559–568, 1984.
24. Misra, A. L. (New York State Division of Substance Abuse, Research Laboratory, 80 Hanson Place, Brooklyn, NY). Personal communication, September 4, 1986.
25. Mule, S. J. The pharmacodynamics of cocaine abuse. *Psychiatr Ann* **14:** 724–727, 1984.
26. Price, H. L. and R. D. Dripps. General anesthetics. I. Gas Anesthetics: Nitrous Oxide, Ethylene, and Cyclopropane. In: *The Pharmacological Basis of Therapeutics, 4th edition,* edited by L. S. Goodman and A. Gilman. London: The MacMillan Company, 1970, pp. 79–82.
27. Pritchard, J. A., R. Mason, M. Corley and S. Pritchard. Genesis of severe placental abruption. *Am J Obstet Gynecol* **22:** 108, 1970.
28. Ritchie, J. M. and N. M. Greene. Local anesthetics. In: *The Pharmacological Basis of Therapeutics, 6th edition,* chapter 15, edited by A. G. Goodman, L. S. Goodman and A. Gilman, New York: MacMillan Publishing Co., 1980, pp. 300–320.
29. Shah, N. S., D. A. May and J. D. Yates. Disposition of levo-[^{3}H]-cocaine in pregnant and nonpregnant mice. *Toxicol Appl Pharmacol* **53:** 279–284, 1980.
30. Stripling, J. S. and E. H. Ellinwood, Jr. Sensitization to cocaine following chronic administration in the rat. In: *Cocaine and Other Stimulants,* edited by E. H. Ellinwood, Jr. and M. M. Kilbey. New York: Plenum Press, 1977, pp. 327–351.
31. Trauner, D. A. and F. L. Mannino. Neurodevelopmental outcome after neonatal cerebrovascular accident. *J Pediatr* **108:** 459–461, 1986.
32. Tuchmann-Duplessis, H. *Drug Effects on the Fetus.* Sydney: ADIS Press, 1975.
33. Verebey, K. and M. S. Gold. Psychopharmacology of cocaine: Behavior, neurophysiology, neurochemistry and proposed treatment. In: *Psycho-Pharmacology: Impact on Clinical Psychiatry,* edited by D. W. Morgan. St. Louis: Ishiyaku Euro-America, Inc., 1985.
34. Vorhees, C. V. Fetal anticonvulsant syndrome in rats: Dose- and period response relationships of prenatal diphenylhydantoin trimethadione and phenobarbital exposure on the structural and functional development of the offspring. *J Pharmacol Exp Ther* **227:** 274–287, 1983.
35. Vorhees, C. V. Behavioral teratology test as a method of screening for hazards to human health: A methodological proposal. *Neurobehav Toxicol Teratol* **5:** 469–474, 1983.
36. Vorhees, C. V., J. Adams, B. K. Nelson, W. J. Pizzi, L. P. Spear, C. F. Mactutus, R. K. Miller, R. E. Butcher, R. Infurna, R. P. Jensh and L. D. Middaugh. *Response of the Behavioral Teratology Society to the Environmental Protection Agency on EPA Test Rule on Triethylene Glycol Monomethyl/Monoethyl/Monobutyl Ethers.* August 7, 1986.
37. Woods, J. R., M. A. Plessinger and K. E. Clark. Effect of cocaine on uterine blood flow and fetal oxygenation. *J Am Med Assoc* **257:** 957–961, 1987.