placed on a diet containing 0, 0.125, 0.25 or 0.50% of lead. Mating was discontinued during week-end. A total of 50 females were used in each group. The mice were dissected on day 16–18 following observation of the vaginal plug, and the number of corpora lutea and of dead and live embryos was recorded. The weight of the live embryos was also determined. Loss prior to implantation is represented by the difference between the number of corpora lutea and that of implanted (live + dead) embryos. The results of the experiments were evaluated by the χ^2 or the t-test.

Results and discussion. The data shown in Table I demonstrate that lead treatment reduces significantly the incidence of pregnancies (mother with at least 1 implant) among females exhibiting vaginal plug. This effect becomes noticeable from doses of 0.25% of lead upwards. Among such pregnant females, the following effects of lead can be noted (Tables I and II): 1. Lead treatment increases significantly the number of embryos dying after implantation, an effect being conspicuous only for 0.5% of lead. 2. The number of embryos dying before implantation decreases after lead treatment with doses of 0.25% or more, so that the total number of embryos implanted per pregnant female increases slightly when related to the number of corpora lutea found. 3. A difference in the weight of live embryos becomes apparent during the last days of pregnancy. This effect is stronger for doses of 0.5% of lead. 4. No gross abnormalities were seen in the lead-treated embryos. This however, does not exclude the possibility that more subtile malformations were present, a problem which will be the object of further study.

Our data confirm the observations of LÉONARD et al.[5] in the mouse, but are at variance with those of SCHROEDER and MITCHENER[3], in that an effect of lead on fertility is seen only after rather high doses. The data obtained suggest that two types of effects of lead on fertility should be distinguished, those on the embryo and those on the mother.

The action on the embryo involves retardation in growth and eventually death at an advanced stage of pregnancy, as shown by the decrease in weight of surviving embryos and the increase in number of embryos dying after implantation. The increase in the number of animals with vaginal plug but no implant could result from the action upon the mother, diminishing her ability to maintain pregnancy. This elimination of the most sensitive females could explain that in the other females (with at least one implant), the preimplantation loss diminishes when compared to that of the controls.

Further studies must show whether this failure is due to a hormonal deficiency, an abnormal placenta or other factors[12].

Summary. Female mice which displayed a vaginal plug after mating were given a diet containing 0, 0.125, 0.250, 0.500% of lead as lead acetate and were dissected 16 to 18 days later. Lead treatment was found to reduce significantly the incidence of pregnancies and to increase the postimplantation loss in the pregnant females.

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Effect of Ethanol Intake on Phenytoin Metabolism in Volunteers

Phenytoin is one of the major antiepileptic drugs. Knowledge about the influence of ethanol intake on its metabolism would be useful for clinical management of the patient with epilepsy. In chronic alcoholics, elimination of phenytoin from plasma after oral dosage is significantly faster as compared with controls. Reduced absorption and induction of enzymes hydroxylating phenytoin have been implicated[1].

It is not known whether acute ethanol ingestion influences phenytoin metabolism in man. It is the purpose of this study to obtain information in healthy volunteers about the effect of acute ethanol intake on the elimination of phenytoin from plasma after a single intravenous dose.

Methods. The 3 female and 2 male volunteers, aged 22 to 47 years, had taken no drugs or alcohol during the 2 preceding months, and were considered healthy according to clinical history and routine laboratory tests. A single i.v. dose of 3 mg/kg phenytoin was injected slowly over 10 min. This was repeated after 7 weeks. At this time, the subjects were administered 1 g/kg ethanol in 40% rum in a sweet drink within 2 h after i.v. injection of 3 mg/kg phenytoin. Plasma samples were taken before and 2, 4, 8, 12, 24 h after the injection of phenytoin. Phenytoin[2] and ethanol[3] plasma concentrations were determined by gas-liquid chromatography in double determinations. The decline of the log phenytoin concentration in plasma with respect to time appeared linear about 4 to 8 h after injection in all subjects. The half-life ($t_{1/2}$) was calculated from the linear part of the curve by the method of least squares. Statistical analysis compared the slope and the elevation of the regression lines in each subject with and without ethanol present in blood.

Results. The average half-life of phenytoin was 12.4 h (SD $\pm$ 4.4) in our volunteers with no ethanol in their blood. After ethanol ingestion, the average half-life of phenytoin was 12.3 h (SD $\pm$ 5.2). The half-life of phenytoin in each subject with and without ethanol in his system was not statistically different at the 5% level (Table). 10 min after the i.v. injection, the phenytoin concentrations ranged from 2.7 to 7.5 µg/ml in the 5 subjects. Blood alcohol levels reached peak values of 0.53–0.80‰ about 1 h after the last intake of alcohol and disappeared linearly with time.

Discussion. The elimination of phenytoin from plasma after a single i.v. dose was monoexponential from about 4 to 8 h after drug administration in all subjects, a finding consistent with apparent first-order elimination kinetics. Ethanol intake with blood levels in a range of 0.53 to 0.80‰ did not influence the half-life of phenytoin in our volunteers. The half-life in the same subject with no ethanol in his blood, determined several weeks before

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The lack of effect of ethanol ingestion on phenytoin half-life

Subject	No ethanol in blood $t_{1/2}$ (h)	Ethanol in blood $t_{1/2}$ (h)	
H.K.	12.0	13.5	n.s.
I.E.	6.3	4.3	n.s.
S.Y.	15.0	14.0	n.s.
D.S.	18.0	18.5	n.s.
I.B.	10.9	11.0	n.s.
	12.4 ± 4.4[a]	12.3 ± 5.2[a]	

[a] Mean ± SD. Half-life of phenytoin was determined from regression analysis of the linear part of the curve of log phenytoin concentration versus time in each subject. The regression lines were compared for slope and elevation in each subject with and without ethanol in blood. The difference was not significant at the 5% level for each subject.

administration of alcohol, served as control. The half-life of phenytoin determined in this study is in very good agreement with values reported recently[4]. The lack of effect of acute ethanol ingestion on the elimination of phenytoin from plasma may be due to two factors. The dose of phenytoin may have been too low to allow for inhibition of phenytoin metabolism to occur with concurrent administration of alcohol. In addition, the blood alcohol level also may have been too low or not been maintained long enough for induction of enzymes hydroxylating phenytoin.

In chronic alcoholic patients, the phenytoin half-life was found to be 16.3 h (SD ± 6.8) as compared to 23.5 h (SD ± 11) in a non-alcoholic control group[1]. In another investigation, 2.5 g/kg alcohol was given daily to 3 volunteers who had stable phenytoin levels[5]. No change occurred in 1 volunteer, in the second a decline, and in the 3rd a rise in the phenytoin plasma levels was seen.

The effect of ethanol on phenytoin metabolism appears complex, since it may either induce or inhibit the enzyme systems hydroxylating phenytoin. The resulting effect may depend on a balance of these antagonistic factors in a subject. A similar experience was made for the effect of ethanol on meprobamate metabolism in man[6].

Summary. Ingestion of ethanol, 1 g/kg, did not influence the phenytoin half-life in 5 volunteers after single i.v. administration of 3 mg/kg phenytoin. The control phenytoin half-life was 12.4 h (SD ± 4.4); with ethanol ingestion it was 12.3 h (SD ± 5.2).

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Variations of Cyclic Nucleotide Monophosphate Levels During Spontaneous Uterine Contractions

There is evidence suggesting that the effect of some smooth muscle relaxing drugs, β-adrenoceptor agonists and phosphodiesterase inhibitors, in several kinds of smooth muscle are mediated by cyclic AMP[1,2]. Contractions following stimulation of cholinergic receptors in intestinal muscle or α-adrenoceptors in vascular muscle have been reported to be associated with an initial reduction of the cyclic AMP level[1]. Smooth muscle contractions have also been found to be combined with an increase of the cyclic GMP level following stimulation of cholinergic receptors in intestine[3] and uterus[4]. The question may be raised whether corresponding changes of cyclic nucleotide levels occur during spontaneous rhythmic contractions and relaxations of smooth muscle. To investigate this problem, we have studied isolated rat uteri.

Uteri from rats weighing 200–250 g were dissected out and the horns were mounted in holders for recording of isometric tension. The rats had been injected with estradiol valerianate (100 µg/kg) or progesterone (4 mg/kg) s.c. for 1–3 days before sacrifice. The preparations were suspended in buffer solution (composition in m*M*) NaCl, 118.5; KCl, 4.7; $CaCl_2$, 2.5; $MgCl_2$, 1.2; $NaHCO_2$, 23.8; KH_2PO_4, 1.2; glucose 5.5 at 37 °C and resting tension was adjusted to about 0.3 g. The solution was oxygenated by 95% O_2 + 5% CO_2. The muscles were equilibrated for about 1 h. At the end of the equilibration period, the spontaneous contractions had become regular. The duration of the contractions of estrogen-dominated uterus was usually about 30 sec, the frequency 1/min and the amplitude 2.7 ± 0.2 g. By the muscle-holders used in this experiment, the preparations could be instantly fixed during various stages of the contraction cycle. The freezing medium used for the fixation was frigen 12, cooled with dry ice (−70 °C). The frozen tissue was extracted with ice-cold 5% PCA. The two cyclic nucleotides were separated on AG-1-x8 formate (200–400 mesh) columns. After lyophilization the nucleotides were dissolved in sodium acetate and analyzed by the methods of GILMAN[5] and STEINER et al.[6] for the respectively nucleotides. Recoveries were usually 85–90% for cyclic AMP and 90–95% for cyclic GMP.

In a recent paper it was demonstrated that cyclic nucleotide concentrations varied during different stage of the estrus cycle[7]. In this study we have also found an effect of estrogen on cyclic GMP level. 20 h after injection

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