acidic histamine-diphosphate used in those experiments. This was found to be the case, since in experiments in which the pH was maintained at 8.4, histamine did not inhibit either the action of diphenhydramine on rat, or of diphenhydramine and of promethazine on guinea-pig mast cells. These results show that the inhibitory effect of histamine on mast cell damage by antihistamines was probably due to a lowering of the pH of the experimental fluid. Histamine by itself does not seem to inhibit mast cell damage induced by antihistamines[5].

Resumen. Se demuestra que la acción de antihistamínicos sobre los mastocitos de la rata y del cobayo depende del pH. Los antihistamínicos estudiados fueron activos en pH 8.4, pero fueron totalmente inhibidos cuando se disminuyó el pH. También se verificó que la histamina no antagoniza la acción de los antihistamínicos sobre los mastocitos cuando el pH fue mantenido en 8.4.

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The Synteratogenic Effect of Lead and Cadmium

The increasing importance of environmental pollution with heavy metals such as lead[1] and cadmium[2] should alert us to the possible effects of these metals on mammalian reproduction. Previous experimental data relating to the effect of heavy metals upon embryonic development in the pregnant hamster have revealed a striking site-specific teratogenic effect for both lead[3] and cadmium[4]. The i.v. injection of cadmium sulfate causes a high incidence of facial abnormalities and a few other malformations including exencephaly and anophthalmia. Under identical experimental conditions the teratogenic effect of various lead salts has been mainly confined to the developing tail bud and associated caudal vertebrae.

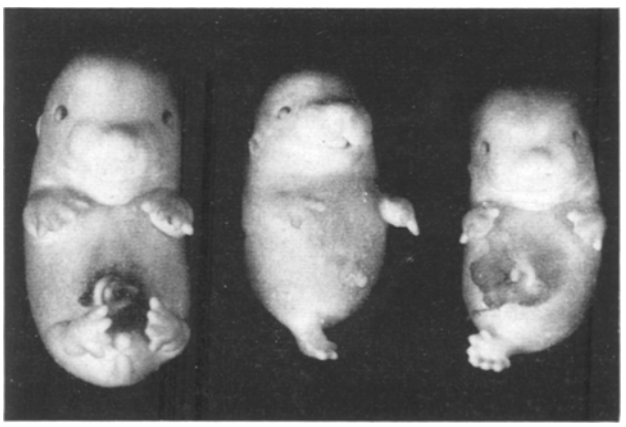
In the present experiments an attempt has been made to combine the teratogenic stimuli of cadmium and lead in order to produce a combination of congenital defects which would reflect the separate teratogenic actions of these agents on diverse parts of the developing embryo. The results were quite different than expected and represent an extremely interesting example of the complex interaction of teratogenic agents.

Female hamsters were bred under direct observation during the early evening hours. The day following the evening of breeding was designated as the first day (day 1) of gestation. On the morning of the eighth day of gestation the animals were anesthetized with pentobarbital and injected i.v. with distilled water (controls), cadmium sulfate, lead acetate or combinations of cadmium sulfate and lead acetate in the amounts and combinations shown in Table I. 4 or 5 days later, on the twelfth or thirteenth day of pregnancy, the maternal animals were killed and the embryos recovered. These embryos were examined carefully for gross external malformations. The number of resorption sites were counted and recorded.

The teratogenic effects of both cadmium and lead when injected separately into pregnant hamsters corresponded well with previous data in that cadmium caused anterior malformations[4] (exencephaly, cleft lip/palate, microphthalmia) only, while lead caused tail malformations only[3]. The combination of these agents, however, revealed that the frequency and severity of the clefts in the lip and palate caused by cadmium are reduced in the presence of lead, while the posterior tail malformations caused by lead appear to be potentiated in the presence of cadmium. Sympodia, a severe caudal malformation of the lower extremities (Figure) was never seen in the animals treated with lead only but did appear with a relatively high frequency when cadmium was added to the lead (Table II).

The teratogenic interaction of other agents has been demonstrated for insulin and 2-deoxy-D-glucose[5] but not for any heavy metals. One can speculate that the teratogenic effect of either of these metals may be due to a direct effect on embryonic tissues, a block in placental transfer of some essential metabolite, or an induced defect in maternal metabolism which secondarily affects the differentiating embryonic tissue.

One obvious possibility which bears further investigation is the well-known importance of heavy metals in the function of several metallo-enzymes which may have



13-day-old fetal hamsters. Animal at left is from a control animal and is normal. The other 2 are littermates from a mother treated with cadmium sulfate (2 mg/kg) and lead acetate (50 mg/kg) on the eighth day of gestation. Both show the same degree of sympodia. 4 other littermates had the same defect. × 3.

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Table I. Effect of cadmium, lead and cadmium-lead on embryonic development in the hamster

	No. of mothers	Total No. of embryos	Normal	Total embryos resorbed	malformed
Cd (2 mg/kg)	7	76	43	21 (27%)	32 (42%)
Cd (2 mg/kg) + Pb (25 mg/kg)	9	122	30	40 (32%)	52 (42%)
Cd (2 mg/kg) + Pb (50 mg/kg)	8	86	0	40 (46%)	46 (54%)
Pb (50 mg/kg)	8	98	14	37 (38%)	47 (48%)
Controls (distilled water)	10	131	121	10 (7%)	0 (0%)

Table II. Malformations resulting from cadmium, lead and cadmium-lead in the hamster

	Exencephaly	Microophthalmia	Cleft lip/palate	Umbilical hernia	Tail
Cd (2 mg/kg)	13	5	19	0	0
Cd (2 mg/kg) + Pb (25 mg/kg)	20	7	8	10	30
Cd (2 mg/kg) + Pb (50 mg/kg)	9	1	0	3	65 (12[a])
Pb (50 mg/kg)	0	0	0	0	40

[a] Sympodia.

important roles in embryonic differentiation. Thus it is possible that under these experimental conditions cadmium and lead interact additively on certain enzyme systems in the case of the tail bud malformation, and that lead blocks the effect of cadmium on the differentiating visceral arch system preventing the facial abnormalities.

Further investigations on the permeability of the mammalian placenta to heavy metals and their localization in specific differentiating embryonic tissues are necessary to identify the exact mechanisms of these site-specific malformations as well as the complex interaction of these teratogenic agents[6].

Zusammenfassung. Bleisalze verursachten fötale Missbildungen der Kaudalregion, wenn sie trächtigen Goldhamstern am 8. Tage injiziert wurden. Kadmiumsalze hingegen verursachten hauptsächlich Gesichts-, Augen- und Gehirndefekte. Die hier beschriebenen Befunde zeigen, dass die Kombination dieser beiden Teratogene zu wesentlich reduzierten Gesichtsmissbildungen, aber erheblich potenzierten Kaudalskelettmissbildungen (Sympus) führt.

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Reversible Necrosis at the End-Plate Region in Striated Muscles of the Rat Poisoned with Cholinesterase Inhibitors

During a systematic study of the processes involved in spontaneous recovery of the rat after severe poisoning with cholinesterase inhibitors, morphological changes in the striated muscles were observed which hitherto remained unnoticed.

Materials and methods. Albino rats of 180–220 g were i.v. injected with a just sublethal dose of either DFP, tabun or paraoxon. The doses used were 1200, 110 and 450 µg/kg, respectively. At suitable intervals after the injection, animals were sacrificed and the diaphragm, the intercostal, the psoas, the gastrocnemius and the soleus muscles were examined histologically. 7 µ thick sections were stained with hematoxilin-eosin. If the sciatic nerve had to be stimulated, the rats were atropinized (50 mg/kg, i.p.) and kept under barbital anaesthesia. If also D-tubocurarine had to be given, the animals were kept alive with artificial respiration.

Results and discussion. About 2 h after the injection of the anticholinesterase localized eosinophilia, swelling of the sarcoplasm and loss of striations is seen in several muscle fibres. In an affected fibre a distinct demarcation exists between the abnormal and the normal parts. After 4–6 h segmented leucocytes appear, the sarcolemnal nuclei become pycnotic and the sarcoplasm breaks up in floes (Figure 1a). Approximately 12 h after the injection, a complete but localized necrosis has developed in the affected fibres. Subsequently histiocytes enter the necrotic parts and phagocytosis starts (Figure 1b). After 2–3 days the remnants of the necrotic sarcoplasm are completely removed and mesenchymal cells proliferate in the apparently empty tubes. Fibrils with striations appear 3–4 days after the anticholinesterase injection. After 10 days the previously necrotic parts are still recognizable as basophilic segments rich in nuclei, but the fibre unity is restored (Figure 1c).

The localized fibre necrosis was the more extensive the greater the activity of the muscle had been, the diaphragm showing more necrosis than the gastrocnemius and psoas muscles. Unilateral section of the phrenic nerve in 6 rats, before the injection of the poison, prevented the necrosis on that side. Unilateral stimulation of the sciatic nerve with one stimulus per 5 sec during 6 h in 5 anaesthetized, DFP-treated rats produced extensive necrosis on that side only.

Strikingly, the anticholinesterase soman, which does not attack the junctional transmission in the rat at a just sublethal dose, produced no necrosis.

Rats treated with the cholinesterase reactivating oxime pralidoxime (P2S), within 2 h after DFP poisoning, showed no necrosis. If the oxime injection was postponed, the necrosis developed as usual.

Direct observation of dissected, living diaphragms under the low-power microscope showed that muscles with necrosis have dark areas in the middle part of the