Detoxification of Cottonseed Pigment Glands with Ferrous Sulfate. (17462)

EDWARD EAGLE (Introduced by A. J. Carlson)

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The use of iron salts as antidotes for cottonseed meal injury was suggested many years ago by Withers *et al.*[1] who successfully fed ferric ammonium citrate-treated cottonseed meal to rabbits and ferrous sulfate and ferric chloride-treated meal to pigs. In a recent paper[2] we studied the acute oral toxicity on rats, mice, rabbits and guinea pigs of cottonseed pigment glands, distinct morphological structures recently made available from cottonseed kernels by a flotation process.[3] The present report concerns LD50 studies on rats of different samples of pigment glands each of which was administered in both distilled water and in 2% ferrous sulfate solution.

Procedure. Male rats weighing between 150 and 220 g were fasted for 18 hours (average weight loss from fasting was 14 g). Water was given *ad libitum*. Each rat was kept in a separate cage in an air-conditioned animal room maintained at 80° ± 1°F and ca. 45% relative humidity. For each of the 3 samples the test preparations were made both by mixing 10 g of the pigment glands with distilled water to a final volume of 100 ml, and by suspending 10 g of the same glands in 2% ferrous sulfate solution to a final volume of 100 ml. Single doses were given by stomach tube on the basis of milligrams of pigment glands per kilogram body weight. All animals had free access to stock diet and water after dosing. Calculations of the LD50

TABLE I.
Mortality in Rats after Oral Administration of Single Doses of Cottonseed Pigment Glands in Water and in Ferrous Sulfate Solution.

	Sample 1		Sample 2		Sample 3		
Dose, mg/kg	In water	In 2% $FeSO_4$	In water	In 2% $FeSO_4$	In water	In 2% $FeSO_4$	In 0.5% $FeSO_4$
7000		0/4		0/4		0/1	4/4
6000		0/4		0/4		0/4	4/4
5400				0/4		0/4	
5000		0/4		0/4		0/4	4/4
4600					4/4	0/4	
4200					5/7	0/4	
4000		0/4		0/4	3/4		4/4
3800					6/7	0/4	
3400		1/4		0/4	4/4	0/5	4/4
3000		0/4		0/4	8/11	0/5	4/4
2600	3/3	0/4	4/4		3/3		
2400			4/4				
2200	3/4	0/4	4/4		6/11		
2000			4/4				
1800	7/7	0/4	3/4		4/4		
1600			3/4				
1400	7/8		2/4		0/8		
1200	2/4		0/4				
1000	3/7						
Total rats used	33	36	32	28	63	36	24
LD50 (mg/kg)	1140	>7000	1490	>7000	2290	>7000	<3000

1 Withers, W. A., and Brewster, J. F., *J. Biol. Chem.*, 1913, **15**, 161; Withers, W. A., and Carruth, F. E., *ibid.*, 1917, **32**, 245.

2 Eagle, E., Castillon, L. E., Hall, C. M., and Boatner, C. H., *Arch. Biochem.*, 1948, **18**, 271.

3 Boatner, C. H., and Hall, C. M., *Oil and Soap*, 1946, **23**, 123; Vix, H. L. E., Spadaro, J. J., Westbrook, R. D., Croveto, A. J., Pollard, E. F., and Gastrock, E. A., *J. Am. Oil Chemists' Soc.*, 1947, **24**, 228.

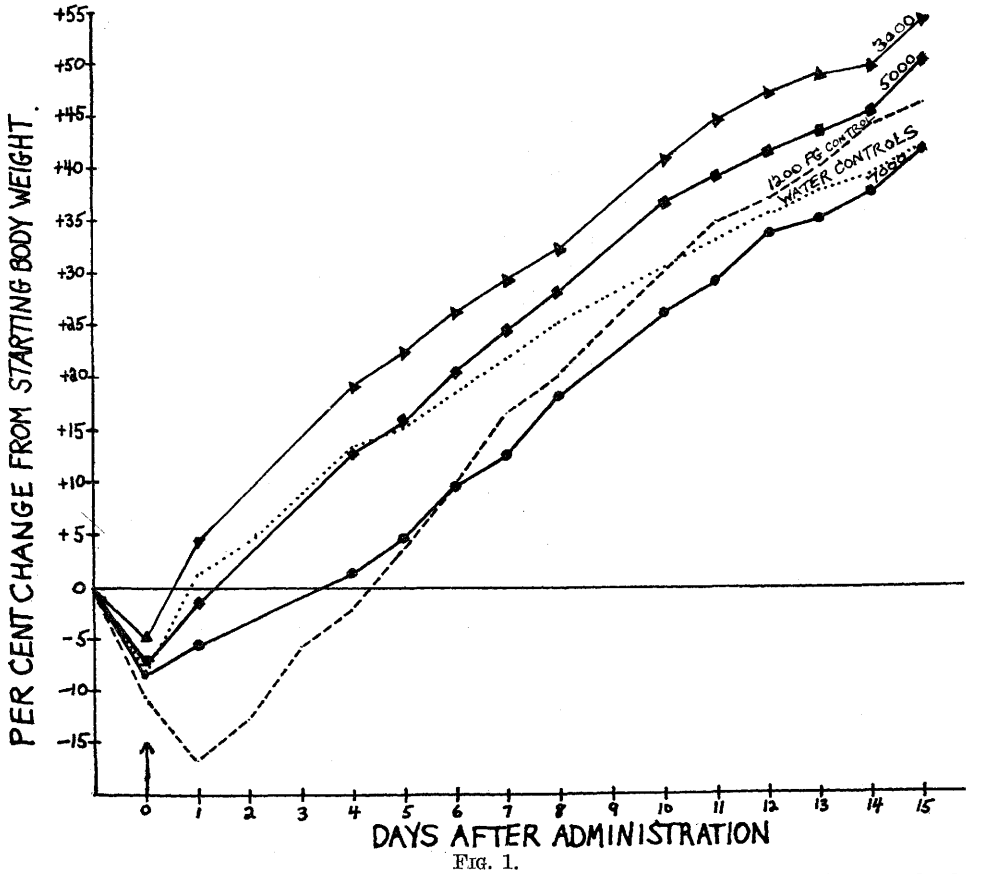

FIG. 1.

Effect on the body weight of the rat of large, single doses of cottonseed pigment glands detoxified by administration in 2% ferrous sulfate solution.

values were made by the method of Reed and Muench.[4]

Results. A summary of the experimental data is given in Table I. Doses of all 3 samples of cottonseed pigment glands which caused 100% mortality when administered in distilled water had no harmful effect when given in 2% ferrous sulfate solution. In the case of Sample 3, cottonseed pigment glands which were completely detoxified when given in 2% $FeSO_4$ lost none of their toxicity when suspended in 0.5% $FeSO_4$ and all of the rats died. The rapid onset of diarrhea reported for cottonseed pigment glands[2] occurred at every dose level when distilled water was the vehicle, but no diarrhea occurred when the glands were suspended in 2% ferrous sulfate solution. All fatalities except one occurred within 3 days, 82% within 24 hours. Typical body weight curves obtained on rats after administration of unusually large single doses (indicated by arrow) of cottonseed pigment glands (Sample 2) are illustrated in Fig. 1. Each point (solid lines) represents an average value obtained from 4 rats. The water control rats (dotted line) received water only; the pigment gland control rats (dashes) received 1200 mg/kg of the glands suspended in water. Since the LD50 value for these pigment glands was 1490 mg/kg, all of the experimental rats would have died had these large doses (3000 to 7000 mg/kg) been administered in distilled water instead of in 2% $FeSO_4$ solution. The 1200 mg/kg dose of pigment glands in water caused a marked depression in body weight; the large doses administered in 2% $FeSO_4$ caused no decreased weight beyond the

[4] Reed, L. J., and Muench, H., *Am. J. Hygiene*, 1938, **27**, 493.

fasting level. Similar body weight curves were obtained with 22 other dose levels of the 3 samples of cottonseed pigment glands.

Summary. Acute toxicity studies were made on 3 different samples of cottonseed pigment glands administered orally to 228 rats in the form of slurries made with either distilled water or 2% ferrous sulfate solution. The LD50 values for the samples when administered in water were, respectively, 1140, 1490 and 2290 mg/kg. When given in 2% ferrous sulfate solution the samples were detoxified so that even such high doses as 7000 mg/kg for each were no longer toxic. There was little effect on the body weight of all 99 rats which survived doses of ferrous sulfate-treated cottonseed pigment glands 3 to 6 times the LD50 value for water-suspended glands. Administration of pigment glands in 0.5% $FeSO_4$ did not detoxify them, and led to the death of all 24 rats given this preparation.

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Effect of 3-Ortho-toloxy-1, 2-propanediol (Tolserol, Myanesin) upon Electrogram of Human Cortex and Subcortex. (17463)

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The sites of action of 3-ortho-toloxy-1,2-propanediol(tolserol, myanesin) have not been exactly determined as yet, although there is suggestive evidence that this drug chiefly affects subcortical ganglia (Berger and Bradley;[1] Burke and Linegar;[2] Davison;[3] Pugh and Enderby;[4] Schlesinger *et al.*[5]). Particularly the experience that it is able to abolish petit mal discharges (Gammon and Churchill[6]) points to a thalamic effect, since these discharges seem to originate in thalamic nuclei (animal experiments of Jasper,[7] human records of Wycis, Lee and Spiegel[8]).

A study of the effect upon the electrical discharges of subcortical ganglia promised to supply more direct evidence. In the course of thalamotomy and mesencephalotomy operations carried out by means of our stereoencephalotome (Spiegel and Wycis[9]), we had an opportunity to record the potentials of human diencephalic and mesencephalic structures preceding the production of the lesions.

The effect of tolserol upon the electroencephalogram (scalp electrodes in the frontal and in the occipital region), upon the electrical discharges of thalamic nuclei (dorsomedial nucleus; lateral nucleus), of the hypothalamus, and of the tectum of the midbrain was studied. Needle electrodes consisting of an inner stylet, insulated except for the tip, and an outer sheath insulated except for a ring 3.0 mm above the tip of the stylet were inserted by means of the stereoencephalotome. Monopolar as well as bipolar leads were used. For monopolar recording, the tip of the inner

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