that this enzyme may have a specific rôle in fatty acid transport.

Summary. Clearing factor lipase, an enzyme with the ability to clear a lipemic substrate and to cause hydrolysis of triglyceride, is released during a single passage of heparin through the vascular beds of the dog liver, dog hindlimb and human forearm.

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Effect of Reserpine and Guanethedine on Excretion of 3-Methoxy-4-Hydroxymandelic Acid in Man.* (28599)

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Rapid depletion of catecholamine tissue stores follows parenteral administration of large doses of reserpine and guanethedine in animals. The mechanism by which this depletion occurs is unsettled. Depletion of catecholamine stores in man following doses employed therapeutically has not been proven. 3-methoxy-4-hydroxymandelic acid (VMA) has been shown to be a specific and major metabolic product of norepinephrine (NE) and epinephrine (E). VMA, which can be precisely measured, appears in the urine in quantities 50 to 100 times greater than NE plus E.

Large intravenous doses of reserpine and guanethedine were therefore administered to human subjects and urinary VMA was measured in order to 1) seek evidence for or against release of catecholamines from body depots and subsequent increased excretion; and 2) to compare the effects of the two drugs.

Methods. Thirty-two subjects were selected, of which 16 were normal volunteers and 16 had uncomplicated essential hypertension established by appropriate studies. One-half were women and half were men. Since no difference in VMA excretion in response to reserpine and guanethedine was found between normotensive and hypertensive subjects, the data from these groups were combined, and 16 whose urine was collected at 8 hour intervals are presented here. No medication had been taken for at least 6 weeks prior to study. During the investigative period, all subjects were placed on a diet containing no fruit, coffee, tea, vanilla or chocolate. The experimental design is shown in Table I. All drugs and placebos were infused during a 15 minute period between 8 and 9 AM. During the infusion and for 45 minutes subsequently, blood pressure was taken every 2 minutes, followed by measurements every 4 hours for the next 23 hours. The cuff method was used. After measurement of urinary pH and volume, creatinine was estimated by means of automatic chemical analyses(1). Aliquots for VMA determination were preserved with a crystal of thymol at −10°C. VMA was determined quantitatively

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TABLE I. Experimental Design.

Drug	No. of subjects	Dosage	Placebo	Urine collection periods
Reserpine	9	70 μg/kg in 300 ml D_5W*	300 ml D_5W	8 AM– 4 PM 4 PM–12 M 12 M – 8 AM
Guanethedine	7	.5 mg/kg in 300 ml D_5W	300 ml D_5W	,,

* 5% dextrose in water.

TABLE II. Effect of Intravenous Reserpine on VMA Excretion.

Quantity	Period	Mean values: Placebo	Mean values: Reserpine	Mean values: Difference	S.E. of difference
μg VMA/kg/hr	8 AM– 4 PM	1.91	1.91	.00	.48
	4 PM–12 M	1.86	1.64	–.22	.63
	12 M – 8 AM	1.24	1.26	.02	.22
μg VMA/mg creatinine	8 AM– 4 PM	3.01	2.80	–.21	.79
	4 PM–12 M	2.16	2.27	.11	.57
	12 M – 8 AM	1.88	2.20	.32	.24
mg creatinine/kg/hr	8 AM– 4 PM	.83	.82	–.01	.14
	4 PM–12 M	.81	.92	.11	.08
	12 M – 8 AM	.74	.66	–.08	.05

TABLE III. Effect of Intravenous Guanethedine on VMA Excretion.

Quantity	Period	Mean values: Placebo	Mean values: Guanethedine	Mean values: Difference	S.E. of difference
μg VMA/kg/hr	8 AM– 4 PM	.90	1.68	.78	.73
	4 PM–12 M	1.05	.88	–.17	.33
	12 M – 8 AM	1.00	.87	–.13	.39
μg VMA/mg creatinine	8 AM– 4 PM	3.73	6.46	2.73	.26
	4 PM–12 M	1.00	1.87	.87	.51
	12 M – 8 AM	1.82	1.25	–.57	.68
mg creatinine/kg/hr	8 AM– 4 PM	.47	.40	–.07	.10
	4 PM–12 M	.64	.55	–.09	.22
	12 M – 8 AM	.69	.69	.00	.17

by the method of Armstrong(2), as modified by Berman and Pettitt(3), using 2-dimensional paper partition chromatography. Reproducibility averaged ± 5%. Recoveries of added VMA, 0.5 to 6 mg, were 95±4%. The basic data used throughout the statistical analysis were the differences between the experimental period and its corresponding control for each subject. Mean differences were analyzed by the "Student's" t-test.

Results. Results are shown in Tables II and III. Neither intravenous infusion of 70 μg/kg of reserpine, averaging 4.5 mg per subject, nor intravenous infusion of 0.5 mg/kg of guanethedine, averaging 31 mg, resulted in a statistically significant change in VMA excretion. VMA excretion values fluctuated within normal limits during both placebo and drug periods in all subjects.

VMA excretion values have been shown both as the rate expression, μg VMA/kg/hr and in terms of μg VMA/mg of creatinine. However, the latter is less accurate since urinary creatinine excretion varies widely in most normal subjects in short collection periods(4), as was the case in this study. The relatively high values for μg VMA/mg creatinine occurring in the 8 AM – 4 PM collection period after guanethedine are due to low creatinine excretions in 2 subjects during this period even though 24 hour creatinine excretions for placebo and drug periods in these 2 patients were equal.

Discussion. The failure of VMA excretion

to increase after maximum intravenous doses of reserpine and guanethedine considered safe for human subjects may indicate that tissue depletion did not occur. Indirect data favoring this possibility as regards guanethedine have recently been published by Cohn(6) who found persistence of pressor responses to ephedrine and tyramine after both acute intravenous and prolonged oral administration in man. If depletion did occur the data reported here are consistent with the hypothesis that this results not from release of catecholamines from tissue stores, but by interference with synthesis of or by blocking storage of these substances.

The measurement of urinary VMA rather than NE and E offers several advantages: 1) more precise measurement is possible; 2) 50-100 times larger amounts for measurement are excreted; and 3) it would be possible more accurately to determine the release from stores if release were taking place slowly enough for enzymatic degradation to keep pace with it.

The possibility that a change in VMA excretion might take place later than 24 hours after drug administration and thus have been missed in the present study must be considered. Against this possibility is the fact that catecholamine tissue levels in animals have always decreased within a matter of a few hours after doses comparable to those used here.

Summary. The effect of intravenous reserpine and guanethedine on urinary excretion of VMA in man has been studied. No remarkable or consistent changes occurred after either drug. Significance of these findings is discussed.

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Effect of Selenium on Muscular Dystrophy in Vitamin E-Deficient Rats and Guinea Pigs.* (28600)

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Schwarz and Foltz(1,2,3,4) have shown that selenium together with Vit. E are among the protective factors against dietary necrotic liver degeneration. Experimental procedures for reproducing this lesion in the rat by feeding diets without Vit. E and deficient in selenium with modifications in quantity and quality of protein, are well known(5,6). Selenium, however, does not prevent all the lesions produced by Vit. E deficiency in different animal species. It prevents exudative diathesis(7,8,9), but not encephalomalacia (10) in Vit. E-deficient chicks. It does not affect the reduced hatching nor high death rate of chickens hatched from the eggs of Torula-fed hens(11), nor sterility due to foetal resorption in rats(12). Selenium prevents multiple lesions in the liver, kidney, heart and skeletal muscle of the mouse on Torula diet(13) and muscular dystrophy in the chick(14), but not muscular dystrophy in the rabbit(15,16) or in the guinea pig(17). Muscular dystrophy in lambs and calves is also prevented and cured by selenite administration. Muth *et al*(18,19) were able to cure with selenite, and not with Vit. E,

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