tic fragility of the erythrocytes of the animals was also examined. The values for the osmotic fragility of thrombocytes in man (Tables I–III) are taken from previous studies[4].

Comment. From the first 3 Tables it may be seen that the osmotic fragility of thrombocytes of the different species varies considerably. Table IV, which gives the results of the osmotic fragility of erythrocytes, indicates similar variations.

Comparison of the figures in the different Tables shows that the osmotic fragility of erythrocytes and thrombocytes is very similar in the same species. The most resistant erythrocytes and thrombocytes were found in man, followed by guinea pig, rat, rabbit, and last by mice and sheep. It is of interest to note that, in the case of resistant thrombocytes like those of man and guinea pig, an intermediate form between normal platelets and platelet ghosts was encountered, namely platelets with sword-like process. In man, where the thrombocytes are most resistant osmotically, the percentage of thrombocytes with a sword-like protrusion in a NaCl solution of 0.30% was 80%, and in the guinea pig only 44%.

The thrombocytes with increased fragility obtained from the other animals, however, showed a direct transition from normal thrombocytes to thrombocyte ghosts, and practically no cells with sword-like processes were encountered.

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Zusammenfassung

Die osmotische Resistenz von Thrombozyten fünf verschiedener Tierarten wurde untersucht und mit der osmotischen Resistenz menschlicher Thrombozyten verglichen. Ein Zusammenhang zwischen der osmotischen Resistenz der Thrombozyten und Erythrozyten der gleichen Tierart konnte gezeigt werden.

Influence of Glutathione on Serum Cholesterol in Rabbits[1]

An increase of intravital oxidation processes could be expected to oxidize cholesterol or its precursors and thus decrease the level of cholesterol in the blood and other tissues. An investigation into whether or not oxidized cholesterol is less atherogenic to animals and whether enhancing *in vivo* oxidation processes decrease the level of cholesterol in the blood of humans and animals, may contribute to the elucidation of these problems. Indeed, Altschul *et al.* could show that

(a) feeding of cholesterol oxidized in various ways prior to giving it to rabbits, produces little or no atherosclerosis[2];

(b) increasing O_2 inhalation tends to decrease serum cholesterol in humans[3];

(c) treating rabbits with pure cholesterol for 3 months and exposing them to increased O_2 tension 3 × weekly, markedly inhibits atherogenesis[4];

(d) ultraviolet irradiation of humans decreases their serum cholesterol[5];

(e) repeated ultraviolet irradiation of rabbits given during the experimental period cholesterol, inhibits atherogenesis[6].

These observations led to the testing of large doses of nicotinic acid which is said to enhance oxygenation of blood and to augment DPN and TPN.

Thus it was found that

(f) nicotinic acid in large oral or parenteral doses lowers serum cholesterol in humans and animals[7] and inhibits atherogenesis in rabbits[8]. The clinical application of this procedure is now widely accepted.

On the other hand, injections of DPN, TPN, catalase, and peroxydase have so far shown no influence on serum cholesterol in rabbits.

Continuing the investigation of substances which may stimulate the activity of respiratory enzymes, Altschul found that

(g) injections of cytochrome C and of hematoporphyrin lower serum cholesterol in rabbits[9].

Recently we have examined the influence on serum cholesterol in rabbits of reduced and oxidized glutathione and of its constituents glycine and cysteine (as DL-cysteine hydrochloride) with the following results.

Methods. Adult male and female rabbits were used. Blood was taken from an ear vein in the morning and the substance to be tested was injected or given in capsule form immediately afterwards. 24, and in some cases 48 h later, blood was again taken. The cholesterol was determined by Schönheimer-Sperry's method as modified by MacArthur. Reduced glutathione was injected intramuscularly in doses of 0.05 g in 16 rabbits, in doses of 0.1 g in 22 rabbits; subcutaneously in doses of 0.1 g in 6 rabbits. It was also given in single capsules of 0.1 g to 6 rabbits and in single capsules of 0.05 g to 6 other rabbits. Oxidized glutathione in doses of 0.1 g was injected intramuscularly in 18 rabbits. DL-cysteine hydrochloride and glycine were each injected intramuscularly into 12 rabbits in doses of 0.1 g. Glycine was also given to 16 rabbits subcutaneously in doses of 0.1 g.

Results. Oxidized glutathione had no significant influence on serum cholesterol by intramuscular or oral administration. Neither had intramuscular injections of DL-cysteine or glycine. Subcutaneous injections of 0.1 g glycine were followed by an average increase in serum cholesterol of 11.9%, but the changes were not regular enough to be significant. Reduced glutathione in capsules of 0.1 g had no effect and neither had intramuscular injections of 0.025 g. Quantities of 0.05 g intramuscularly increased the serum cholesterol after 24 h by 6.6%, but the results were not statistically significant. However, doses of 0.1 g of reduced glutathione, injected intramuscularly, increased serum cholesterol after 24 h to a highly significant degree (see Table). In 19 cases the serum cholesterol was also determined 48 h after the injection of 0.05 g and 0.1 g of reduced glutathione. In 8 of these cases the increase of serum cholesterol surpassed the 24 h increase, in 10 cases the cholesterol values were lower than those after 24 h and in one case the value was unchanged.

[1] This investigation was supported by a research grant H-4568 from the National Heart Institute, Public Health Service (U.S.A.).

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Influence of glutathione 0.1 g (i. m.) on serum cholesterol in rabbits

Rabbit No.	Cholesterol (mg%) before injection	Cholesterol (mg%) 24 h after injection	Change %
1–27	26	34	+30.8
2–29	71	79	+11.3
3–9	18	22	+22.2
4–175	134	158	+17.9
5–8	107	138	+29.0
6–129	35	44	+25.7
7–135	48	49	+ 2.1
8–23	116	135	+16.4
9–51	106	105	− .94
10–88	73	82	+12.3
11–50	65	92	+41.5
12–173	56	54	− 3.6
13–87	39	50	+28.2
14–172	44	63	+43.2
15–45	63	69	+ 9.5
16–178	54	77	+42.6
17–151	42	70	+66.7
18–37	33	33	0
19–1	51	65	+27.5
20–101	106	106	0
21–38	50	67	+34.0
22–150	54	69	+27.8
	Average 63.2	Average 75.5	Mean +21.99%
			S. D. ±17.30
			t = 5.83
			$P <$ 0.001

With regard to the mode of action of reduced glutathione in raising serum cholesterol in rabbits, it seems futile at the present time to attempt an explanation in view of the scanty knowledge about the function of this substance. However it deserves mention that according to LEVY and POPJÁK[10] cysteine and glutathione activate mevalonic kinase which plays a role in converting mevalonate into squalene, both of which are precursors of cholesterol.

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Zusammenfassung

In Kaninchen erfolgt auf intramuskuläre Injektionen von 0.1 g reduziertem Glutathion nach 24 h ein hoch signifikanter Anstieg des Serumcholesterins. Kleinere Dosen, oder orale Verabreichung von 0.1 g, haben keine derartige Wirkung.

Oxydiertes Glutathion, DL-Cystein und Glykokoll zeigen bei Kaninchen keinen eindeutigen und unmittelbaren Einfluss auf Serumcholesterin.

Multiple Effects of Oestrone and Testosterone Propionate in Combination: Simultaneous Effects on Vaginal Keratinization and Uterine Growth

For some years androgens of various types have been known to inhibit the vaginal effects of oestrogens[1]. The recent demonstration that testosterone propionate may augment the action of the important natural oestrogens in stimulating uterine growth[2] suggested the likelihood that the androgen in a single oestrogen-androgen combination might both augment and inhibit the oestrogen simultaneously at different target organs. This could be observed in the same animal. Such a study, employing combinations of oestrone and oestriol, had been suggestive, but not conclusive[1]. The following experiment was designed to determine whether in combination with oestrone, testosterone propionate would appear as a synergist and antagonist respectively at the uterus and vagina of the same animal.

The experimental approach has been described previously[2]. In short, 30-day old Sprague-Dawley rats were spayed and rested 10 days to insure regression of the uterus. Then injections were given daily for the next three days, and the animals were sacrificed 24 h after the final injection. At autopsy the uterus was removed, cleaned of adherent tissues, scored and blotted to remove contained fluids, and weighed wet on a torsion balance. At the same time the vagina was dissected out, fixed, sectioned at 7 μ, stained with hematoxylin and eosin, and studied microscopically. Four groups of 14 rats each were employed. One group received solvent injections, 0.1 ml of corn oil; the second group was injected with 1.0 μg of oestrone each day; the third group, 500 μg of testoterone propionate daily; and the fourth, 1.0 μg of oestrone and 500 μg of testosterone propionate in single daily injections of 0.1 ml of corn oil. (Tab.)

The uteri of the oil-treated animals were atrophic, as was to be expected in spayed animals, but both the oestrone and testosterone propionate separately produced considerable uterine growth. The addition of the androgen to the oestrogen stimulated an additional increment of uterine growth. Thus this test satisfactorily repeated earlier studies demonstrating that oestrone-induced uterine growth may be augmented by testosterone propionate.

Keratinization of the vaginal epithelium, on the other hand, was inhibited by the androgen (Table). None of the

The effects of 1.0 μg of oestrone and 500 μg of testosterone propionate administered daily for three days, alone and in combination, on the uterine growth and vaginal histology of spayed rats

	N	Uterine Weight[a]	Vaginal Indices: Keratinization	Vaginal Indices: Mucification
Oil only	14	*27.9*	0/14	0/14
Testosterone propionate	14	*65.2*	0/14	3/14
Oestrone	14	*83.2*	9/14	0/14
Oestrone + TP	14	124.5	0/14[b]	14/14[c]

[a] Uterine weights for groups designated by italic ciphers differ significantly with $P < 0.01$, by Wilcoxon Rank Sum Test[3].

[b] Significantly different from oestrone-treated group. Chi-Square = 10.48, $P < 0.01$.

[b] Significantly different from testosterone propionate-treated group. Chi-Square = 14.97, $P < 0.01$.

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