Effect of Serum Gonadotropin on Testis in Ascorbic Acid Deficient Guinea-Pigs

Degeneration of both germinal epithelium and Leydig cells have been reported in vitamin C-deficient guinea-pigs[1]. Spermatogenic arrest and an alteration in some of the cytoplasmic constituents in the seminal vesicles have also been observed under similar conditions[2,3]. LLAURODO and EIK-NES[4] have failed to detect any changes in testicular ascorbic acid of either rat or dog on administration of Human chorionic gonadotropin (HCG). In order to have an idea whether the testicular damage as observed in ascorbic acid deficiency is due to reduced output of pituitary gonadotropins, the effect of injection of serum gonadotropin on the scorbutic testis has been investigated.

30 male guinea-pigs of average weight 230–260 g were selected for the experiment. They were divided into three groups of equal number. All the groups received a scorbutic diet[5] daily and a concentrate of vitamin A, D, E and K twice a week. The animals of the control group were fed 5 mg of vitamin C per animal per day in addition. One group of scorbutic animals was injected with serum gonadotropin (Gestyl, 20 IU/100 g body weight) each day from 15 days on a scorbutic diet until the termination of the experiment. The animals of all the groups were paired-fed and killed on the 23rd day of the experiment by cerebral concussion. Testes of the animals were removed and fixed in Carnoy. Paraffin sections were stained with hematoxylin and eosin for histological study. The seminal vesicle and prostate were collected, and weighed in torsion balance.

Spermatogenic inhibition and atrophy of the testes were noticed in all the ascorbic acid deficient and in two of inanition control guinea-pigs. Most of the scorbutic guinea-pigs treated with gonadotropin showed a normal picture of the testes with mature sperm in the majority of the tubules. The atrophy of seminal vesicle and prostate in scorbutic guinea-pigs has also been observed to improve after the hormone treatment (Table). Such treatment, however, did not retard the onset of scurvy in these animals.

The role of ascorbic acid either in spermatogenesis or in biosynthesis of androgenic hormone is not established. NESPOR[6] observed a fall in the ascorbic acid content of the testis of guinea-pigs on scorbutic diet after giving gonadotropic hormone. LLAURADO and EIK-NES[4], however, have reported that increased steroid output by the testis on administration of human chorionic gonadotropin does not cause parallel alteration in the ascorbic acid content of the particular organ. NOACH and VAN REES[7] also observed no alteration in testicular ascorbic acid following repeated administration of either chorionic or serum gonadotropin. On the other hand, in ascorbic acid deficiency spermatogenic arrest and degeneration of interstitial cells have been noted in guinea-pigs[1]. DEB and CHATTERJEE[8] have observed that the testicular degeneration which follows after administration of alloxan can be corrected by a massive dose of ascorbic acid treatment. They have suggested that possibly the degenerative changes observed are due to blood ascorbic acid lowering action of the diabetogenic agent. The mechanism through which ascorbic acid deficiency produces testicular atrophy is not established. Several investigators[9–11] have reported adrenal hyperactivity during scurvy. Inhibition of pituitary gonadotropin following release of an excessive

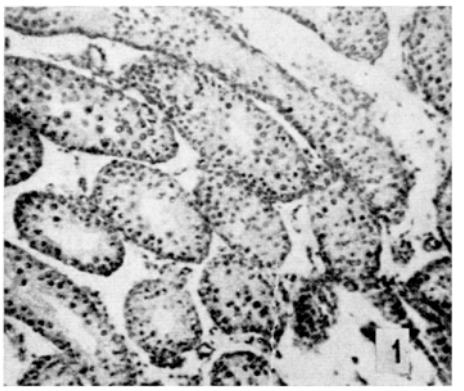

Fig. 1. Testis from scorbutic guinea-pig, showing complete inhibition of spermatogenesis. × 96.

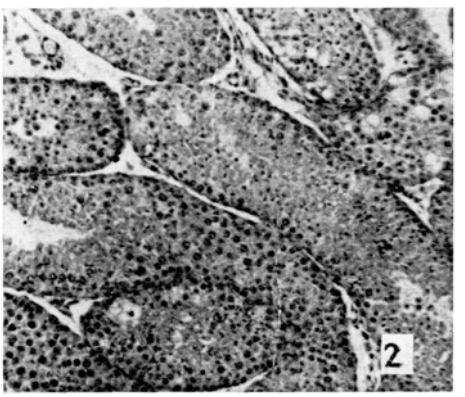

Fig. 2. Testis from scorbutic guinea-pig treated with serum gonadotropin. Normal spermatogenesis can be noted. Compare with Figure 1. × 96.

Effect of serum gonadotropin on weights of accessory sex organs in scorbutic guinea-pigs

	No. of animals	Seminal vesicles (empty) mg/100 g body weight	Prostate mg/100 g body weight
Control	10	94.78 ± 8.60[a]	92.44 ± 4.42
Scorbutic	10	38.38 ± 3.34	36.07 ± 3.31
Scorbutic and Serum gonadotropin treated	10	98.40 ± 2.92	87.66 ± 2.61

[a] Means ± Standard error.

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amount of ACTH have previously been observed[12,13]. BANERJEE and GHOSH[14] have reported the histochemical changes in testes and adrenal glands during scurvy to be corrected on administration of ascorbic acid. DEB and BISWAS[15] have suggested that the inhibition of spermatogenesis in rats produced by high tyrosine diet was possibly due to a reduced secretion of hypophyseal gonadotropin, although they have noted a correction of the said disturbances on vitamin C. The functional activity of the pituitary gland in ascorbic acid deficiency has not been studied in detail. DEB and BANERJEE[16] have noted a fall in alkaline phosphatase in the anterior pituitary during scurvy and postulated it to be possibly due to diminution of some pituitary trophic hormone. Recently, pituitary bioassay revealed that the gonadotropic potency has been increased in C-deficient guinea-pigs (DEB and BISWAS[17]). An increased gonadotropic content and reproductive disturbances also have been reported in B_6-deficient rats[18,19]. The testicular degeneration in ascorbic acid deficiency may, therefore, be caused by failure of the pituitary to release sufficient gonadotropic hormone[20].

Résumé. La dégénération des testicules de cobayes manquant d'acide ascorbique a pu être corrigée par l'administration du sérum gonadotropin.

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Effect of Silica on Protein Biosynthesis by Rat Liver Cell-Free Systems

Silica particles engulfed by macrophages in vitro bring about an injurious effect to these cells. The mechanisms of cytotoxic action of silica and their specificity have been widely investigated in recent years on cultured macrophages and on various cells and tissues by several workers[1–8]. Nevertheless, the nature of the toxic effect of silica remains a problem still open to question.

The present report is a further study[9,10] on the same question. It deals with the action of particulate silica on protein biosynthesis at subcellular levels by investigating the amino acid incorporation rate into protein by rat liver cell-free extracts.

The microsomal system of ZAMECNIK and KELLER[11] as described by STEIN and GROSS[12] has been used, and purified preparations of crystalline HF-treated silica (tridymite) and of vitreous HF-treated silica (both in a diameter range $\sim 1\ \mu$) tested. Liver homogenates 1:3 by volume were prepared with teflon pestle homogenizer in $0.25\,M$ sucrose with $0.025\,M$ $KHCO_3$, $0.02\,M$ potassium phosphate buffer pH 7.8 and $0.01\,M$ nicotinamide. Nuclei and cell debris were removed by centrifugation at 800 *g* for 6 min and the resulting supernate recentrifuged for 15 min at 10,000 *g*. Centrifugations were made in a refrigerated International centrifuge. 1.0 ml of the 10,000 *g* supernate was incubated in Warburg flasks with 20 μM fructose 1-6 diphosphate potassium salt, 2.0 μM ATP potassium salt, 2.0 μM $MgCl_2$ and 0.26 μM DL-leucine-1-C^{14} (specific activity 3.8 $\mu c/\mu M$) in a final volume of 2.0 ml. When present silica was in amount of 15 mg. Incubations were carried out for 1 h at 37°C in a Warburg metabolic shaking incubator with O_2 95% + CO_2 5% as gas phase. The reaction was stopped by TCA and protein

In vitro incorporation of DL-leucine-1-C^{14} into rat liver microsomal protein. Each figure (c.p.m. per mg protein) is the mean ± S.E. of 2 flasks

Experiment	Control	Crystalline silica	Vitreous silica
1	92.5 ± 4.47	52.0 ± 1.25	–
2	80.5 ± 5.48	59.0 ± 5.00	–
3	116.0 ± 3.00	62.0 ± 1.40	–
4	111.0 ± 9.00	65.0 ± 6.00	–
1	81.0 ± 3.00	–	65.5 ± 7.48
2	75.5 ± 10.49	–	43.0 ± 6.00
3	84.0 ± 10.00	–	76.5 ± 3.46
4	80.5 ± 5.48	–	62.5 ± 7.48

The statistical evaluation of data has been made with the analysis of variance test.

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