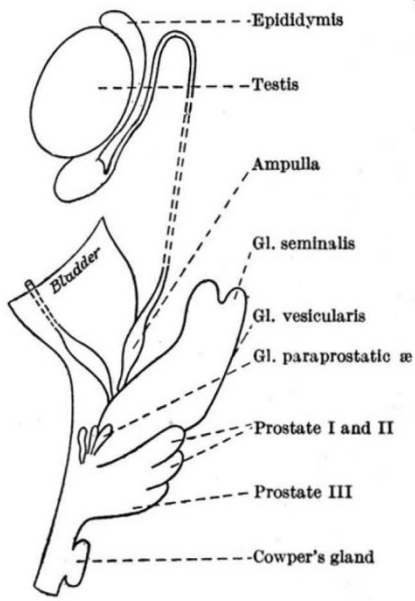
Functional Development of Accessory Glands and Spermatogenesis

WHEN fructose was established as the normal nutrient material for spermatozoa and its origin traced to the seminal vesicles[1], it remained to be explained how certain animals, such as the rabbit, secrete fructose in semen in spite of the absence of seminal vesicles. The rabbit, however, has a large complex organ, sometimes referred to as the 'prostate', which has been described hitherto as composed of three glands, 'gl. seminalis', 'gl. vesicularis' and 'prostate proper', each with an independent urethral outlet[2]. However, our recent investigation of the development of the male reproductive system in the rabbit has established that the gl. seminalis develops in conjunction with the gl. vesicularis from the same diverticulum of the Wolffian duct; further, both glands possess a common urethral outlet, and gl. vesicularis as a whole, rather than the gl. seminalis by itself, should be regarded as homologous to the seminal vesicle in other mammals[3]. This, incidentally, explains the presence of fructose in the gl. vesicularis. It should be pointed out, however, that in the rabbit, apart from this gland, fructose also occurs in the ampulla of the vas deferens as well as in the 'prostate proper'. The distribution of fructose in the various parts of the reproductive system of the male rabbit is shown in the accompanying sketch.



	Weight of organ (mgm.)	Fructose in organ (mgm.)	Fructose (mgm./100 gm. tissue)
Epididymis	600	0·03	5
Testis	2900	0·06	2
Ampulla	130	0·03	23
Gl. seminalis	150	0·04	26
Gl. vesicularis	920	0·79	85
Gl. paraprostatic æ	190	0·092	48
Prostate I and II	335	0·368	110
Prostate III	475	0·272	57
Cowper's gland	1000	0·0	0·0

In the course of investigations on the development of the reproductive system in the rabbit, we noticed that fructose appeared in the accessory glands at an early stage when there was as yet no sign of active spermatogenesis. In a four-months-old animal, both the gl. vesicularis and the prostate showed already a fairly high concentration of fructose (21 and 44 mgm. per cent, respectively) in spite of the complete absence of spermatozoa in the testis or the epididymis. When, in the sixth month of life, the spermatozoa finally made their appearance, the accessory glands were filled with secretory fluid containing the normal high level of fructose. Experiments on bull-calves gave similar results, showing that the appearance of fructose in the secretory fluids of the accessory glands precedes the onset of active spermatogenesis.

Thus it appears that the male reproductive organs first accumulate a store of nutrient material, so that when the motile spermatozoa make their appearance in the generative tract, the fructose reserve is available, ready to be utilized. Together with the recent finding that the formation of seminal fructose requires the presence of the testicular hormone[4], our experiments provide additional evidence that the testicular hormone begins to function in the body some time before actual spermatogenesis.

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[1] Mann, T., *Biochem. J.*, **40**, 481 (1946).
[2] Leydolph, W., *Z. mikr.-anat. Forsch.*, **19**, 285 (1930).
[3] Davies, D. V., and Mann, T., *Proc. Anat. Soc.*, April 25, 1947.
[4] Mann, T., and Parsons, U., see preceding communication.

Oxidation of Insulin by Performic Acid

FROM the determination of the terminal residues of insulin, it was suggested that the submolecule of molecular weight 12,000 is made up of four peptide chains bound together by —S—S— linkages[1] Thus if one could break the —S—S— linkages without affecting any other part of the molecule, it should be possible to split the insulin into its separate polypeptide chains, two of which have terminal glycyl residues and the other two phenylalanyl residues. Toennies and Homiller[2] showed that the only amino-acids that are appreciably oxidized by performic acid are tryptophan, methionine and cystine, the latter reacting with five atoms of oxygen and presumably forming cysteic acid. Since insulin contains no tryptophan or methionine, this seemed a suitable way of splitting the —S—S— linkages.

Using the procedure of Toennies and Homiller, it was found that the oxidation of cystine to cysteic acid is complete in five minutes. With insulin an oxidation-time of 15 minutes was generally used. The oxygen consumption was the theoretical one for the cystine content, and paper chromatography[3] showed no qualitative difference in the amino-acid composition except the replacement of cystine by cysteic acid. There was no destruction of the free amino-groups[1]. The oxidation product was studied in the electrophoresis apparatus of Tiselius. Unfortunately, it was not possible to dialyse the material, due to its low molecular weight (about 3,000), so that the results were not always entirely reproducible. Fig. 1 illustrates a typical experiment, which shows three components and indicates that the mixture is not unduly complex.

Fig. 1. ELECTROPHORETIC DIAGRAM OF OXIDIZED INSULIN

Fig. 2. ELECTROPHORETIC DIAGRAM OF FRACTION *A*