tion may not be noticeable until later in life when the animal has almost attained maturity. The infection is attended by a sudden variation[5] in the virus manifesting itself by its inability to infect adult individuals of the original species; by the sort of cells attacked and characteristics of resultant tumors in the new species; and possibly by changes in its antigenic make-up. Duplicating the same sequence but in an inverse order, infection of the original species by the virus variant is only accomplished in the very young individual, which weeks or months later may develop a neoplastic disease radically different from that induced in it by the original virus. But this back infection is not attended by any other obvious variation in the virus which now attacks young individuals of both species.

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INOSITOL AND SPECTACLED EYE IN RATS

IN the biological assay for vitamin B_6 employing the diet of Halliday and Evans,[1] it was observed that denudation around the eyes could be induced in almost 100 per cent. of the fifteen animals when nicotinic acid was given at weaning in addition to thiamin, flavin, pantothenic acid and choline. This syndrome was similar to that reported by Oleson, Bird, Elvehjem and Hart,[2] except that the spectacled eye was not complicated by the exudation and final closure which Unna[3] reports could be cured by pantothenic acid.

Apparently the spectacled eye, as previously reported by other workers, is a multiple deficiency requiring at least two factors, one of which is pantothenic acid and the other a factor concerned in the growth of hair.

It is known that filtrate factor concentrates of liver, yeast and cereal grains can cure both the denudation around the eyes as well as the exudation condition. Since the true spectacled eye might be thought of as a type of alopecia, and since Woolley[4] has shown the mouse alopecia factor to be inositol, this compound was fed to rats in an attempt to cure the spectacled eye as produced under our conditions. 10 mg of Eastman's inositol were administered per rat per day. The results were quite dramatic. The swelling around the eyes disappeared within 24 hours and in three days definite signs of hair restoration were evident. By 10 to 14 days the halo around the eyes was completely overgrown with hair and the rats could not be distinguished in this respect from normal animals.

In harmony with Woolley's[4] report, a definite response in growth accompanied the above changes. On the Halliday and Evans diet supplemented with crystalline thiamin, flavin, nicotinic acid, pantothenic acid, pyridoxin and choline, these control animals showed an average weekly gain of 10 grams. When 10 mg of inositol were given in addition, the average weekly gain was 15 grams.

We believe the evidence demonstrates inositol to be the factor concerned with the regeneration of hair in the condition referred to as "spectacled eye" in rats. In addition, inositol has been shown to have a function in the growth of the rat.

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[5] The word *mutation* is purposely avoided until an agreement is reached as to whether or not it is permissible to use it in fields other than genetics.

[1] N. Halliday and H. M. Evans, *Jour. Nutrition*, 14: 45, 1937.

[2] J. J. Oleson, H. R. Bird, C. A. Elvehjem and E. B. Hart, *Jour. Biol. Chem.*, 127: 23, 1939.

[3] K. Unna, *Jour. Nutrition*, 20: 565, 1940.

[4] D. W. Woolley, SCIENCE, 92: 384, 1940.

THE DEVELOPMENT OF EARLY MOUSE EMBRYOS IN THE EXTRAEMBRYONIC COELOM OF THE CHICK[1]

IN preparation for an experimental study of the development of certain hereditary abnormalities affecting early embryonic stages in mice, a new method for cultivating entire early mouse embryos has been developed. Mouse embryos at the ages of 7 to 8 days, *i.e.*, egg cylinder to six somite stages, were removed from the uterus within their decidua and transferred into warm sterile Ringer's solution. They were then dissected out of the decidua and Reichert's membrane was pulled off the egg cylinder. For the transplantation of stages older than the egg cylinder all membranes were removed from the embryo as completely as possible. The embryos were transplanted into 72 to 80-hour chick embryos in such a way that they came to lie in the extraembryonic coelom. A window was cut into the egg shell, then a slit was made into the vitelline membrane and the serosa; the mouse embryo was transferred onto the membranes with a Spemann pipette and pushed through the slits into the extraembryonic coelom next to the allantois with the help of glass needles.

At examination after 24 or 48 or 75 hours the mouse embryos may be found floating freely in the extraembryonic coelom, or attached to any of the extraembryonic membranes; in some instances they are attached to the allantoic stalk. Some operations were performed in which the mouse embryo was pushed partly into the coelom at the place of open communication between intraembryonic and extraembryonic coelom next to the allantoic stalk. Such embryos con-

[1] These studies were aided by a grant to Professor L. C. Dunn from the Fund for Research of Columbia University and from the Josiah Macy, Jr., Foundation.

tinued their development at the site of implantation, *i.e.*, partly in the intraembryonic, partly in the extraembryonic coelom; this location was found to be especially suitable for the further development of the mouse embryos. Fourteen primitive streak stages (7 days) transplanted to the chick by the methods described above developed within 48 hours into typical 9-day embryos which were smaller than normal but looked fairly normal, at least externally. However, a disturbance of the size relationship of the different organs was noticed. The embryos were enclosed in their amnion, had six pairs of somites, a strongly and normally beating heart, cerebral lobes which were well developed but relatively smaller than normal, an irregular slightly winding neural tube and a tail bud. Other embryos, implanted into the extraembryonic coelom in the first somite stages (age about 8 days), continued development up to the stage when maternal circulation becomes of primary importance for the nutrition of the embryos, *i.e.*, at 9 to 10 days. Further experiments will have to show whether it will be possible to obtain any development beyond these stages. By observing aseptic precautions, the operations can be made to work successfully without great difficulties in a large percentage of cases.

Thus a method for extended study of post-implantation stages of mammals has been developed which will supplement those already made available by Nicholas and Rudnick[2] and by Törö[3] for rats, and Waddington and Waterman[4] for rabbits.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

AN APPARATUS FOR CONTINUOUS, RAPID AND COMPLETE EXTRACTION OF ESTROGENS

MANY continuous extraction apparatus have been developed for use in estrogen determinations.[1,2,3] In a a series of clinical studies now in progress, where multiple determinations are required, we designed an extractor which permits small quantities of liquid to be rapidly extracted as a part of a routine in this series of analyses.

The apparatus schematically illustrated consists of several separate units which are connected by standard ground glass joints. These are boiling flask (*B*), condensing unit and extractor (*A*), which includes an inlet and outlet to facilitate operation, and a stirring device to increase extracting efficiency.

In flask *A* is placed the solution to be extracted to a level about 2 cms below the opening into tube (*a*). In flask *B* is the extracting solvent which is of a lower specific gravity. When heat is applied to flask *B* vapors developed go up (*b*) are condensed, and dropped down through tube (*c*) to pass out at the bottom of flask *A* through perforated bulb (*d*). The stirrer mixes the two liquids into a temporary suspension without disturbing the layers formed at the top. The lighter liquid collects at the top until the hydraulic head built up in (*c*) is sufficient to cause siphonation at tube (*a*) into flask *B*. From here the whole process repeats itself, continuously. The apparatus is filled through a funnel (*D*) and at the end of the operation is emptied through the stop-cock (*e*) opening at the base of flask *A*.

The inclusion of the stirring device increases extracting efficiency by bringing more liquid into contact with extracting solvent. The inlet funnel (*B*) and outlet stop-cock (*e*) permit filling, separation of layers and emptying without loss of time and danger of breakage in disengaging at the start or conclusion of an operation. This also facilitates cleaning of the apparatus.

This extractor can be adapted for large quantities by substituting a larger extraction flask of relative proportions. The advantage of the stirrer in a larger apparatus becomes more apparent as the peripheral liquid which would ordinarily not be touched by the extracting solvent is brought into direct contact due to stirring action.

Several tests on the efficiency of the apparatus have been made. Specimens used were estrone—estradiol fractions (obtained according to the method of Smith and Smith, 1939, from three separate pooled pregnancy urines, each divided into equal aliquot portions as labeled in Table 1).

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[3] Emeric Törö, *Jour. Exp. Zool.*, 79: 213–236, 1938.
[4] C. H. Waddington and A. J. Waterman, *Jour. Anat.*, 67: 355–370, 1933.