This is in agreement with the findings of Menken[3] who studied the effect of the adrenal cortical substances on the accumulation of trypan blue in local lesions produced by the injection of leukotoxine or inflammatory exudate.

[3] Menken, V., PROC. SOC. EXP. BIOL. AND MED., 1942, **51**, 39.

14321 P

Factors Controlling Lens Regeneration from the Dorsal Iris in the Adult *Triturus viridescens* Eye.*

L. S. STONE.

From the Department of Anatomy, Yale University School of Medicine, New Haven, Conn.

When the adult eye of *Triturus viridescens* is transplanted[1,2] or its blood supply is temporarily lost[3] the lens degenerates along with most of the retina. In these cases a new lens is developed from the dorsal pupillary margin of the iris such as follows simple lensectomy.[3,4] This phenomenon of lens replacement is largely, if not exclusively, confined in the vertebrates to the Triturus group of salamanders[5,6,7] where the lens appears to be less resistant to injury than in other forms.[5,8] An extensive study is being made by the author in search for the factors which govern this phenomenon. Some of the findings can be summarized at this time.

Slight injuries to the fully developed lens in the adult eye of *Triturus viridescens* cause it to degenerate. Injuries to the dorsal iris produced by mechanical stretching and incisions radiating from the pupillary margin neither prevent nor favor lens regeneration. Such injuries heal readily. Only the presence of the normal lens prevents a new one from forming. This was shown by means of a successful pipette method of removing and reimplanting the lens in the adult eye without injury.

If a piece of the dorsal iris from a normal eye or from one lensectomized 6 or 7 days earlier is transplanted to a freshly lensectomized host eye, a new lens will develop from the graft. If, however, such grafts are placed in an eye possessing a normal lens, the latter sharply inhibits lens regeneration in all cases. Even in those grafts where the implanted iris was allowed to initiate early preparation[7] for lens regeneration through delay before transplantation, only a rare case showed at best a feeble, short-lived attempt to progress further before it was permanently arrested. The presence of the normal lens of the host eye apparently exerts an early effect upon the lens-forming capacity of the implanted iris.

Varying the time from the day of operation to 25 days after lensectomy in both host and donor eyes, combinations were obtained which tested the effects of older and younger regenerating lenses on each other in the same environment. The experiments proved that the presence of a regenerating lens from an iris up to 25 days after lensectomy does not inhibit nor prevent the regeneration of a lens from other dorsal iris tissue in its environment. Under some circumstances as many

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[1] Stone, L. S., and Zaur, I. S., *J. Exp. Zool.*, 1940, **85**, 243.

[2] Stone, L. S., and Farthing, T. E., *J. Exp. Zool.*, 1942, **91**, 265.

[3] Stone, L. S., and Chace, R. R., *Anat. Rec.*, 1941, **79**, 333.

[4] Ogawa, C., *J. Exp. Zool.*, 1921, **33**, 395.

[5] Stone, L. S., and Sapir, P., *J. Exp. Zool.*, 1940, **85**, 71.

[6] Stone, L. S., and Dinnean, F. L., *J. Exp. Zool.* 1940, **83**, 95.

[7] Dinnean, F. L., *J. Exp. Zool.*, 1942, **90**, 461.

[8] Stone, L. S., and Cole, C. H., *Yale J. Biol. and Med.*, 1943, **15**, 735.

as 3 lenses may grow in the same environment. However, between 25 and 30 days after lensectomy the regenerating lens demonstrates clearly that at this time it becomes linked with a factor, as in the case of a normal fully grown lens, which acts as an inhibitor for lens regeneration from the iris within the eye. This is at a time when the newly forming lens detaches itself from its source of origin, and probably marks a definite period in its differentiation. Both implantations of the detached lens and implantations of various types of dorsal iris emphasize this point.

The results of the experiments carried out within the eye environment seem to be significant. So far it has been found that the dorsal iris grafts of the adult eye of this species only rarely made a feeble but aborted attempt to regenerate a lens when placed outside of the eye environment such as under the skin or within the body cavity. However, this inhibiting effect is possibly of a different nature than that exerted by the lens itself.

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Concentration and Preservation of Crude Penicillin.

TSUN T'UNG. (Introduced by Martin Frobisher, Jr.)

From the Department of Bacteriology, School of Hygiene and Public Health, Johns Hopkins University.

Rapid advances in methods for the purification of penicillin seem to have rendered the small-scale laboratory production of the crude material undesirable. However, since the processing of pure penicillin is complicated and the yield is very small, general clinical trials and intensive laboratory investigations of this substance will not be possible for some time to come. Therefore, the crude penicillin is still useful, provided its bulk can be reduced without loss of potency. The present communication presents a simple and inexpensive procedure for concentrating and preserving crude penicillin for practical use.

The Fleming strain of *Penicillium notatum*[1] was grown on modified Czapek-Dok medium[2] contained in Blake bottles. In order to minimize precipitation of the constituents of the medium, brown sugar and ferrous sulfate were separately sterilized by autoclaving and filtration through Seitz pad, respectively, and were then added to the basic medium.

After one week of incubation at room temperature the penicillin titer of each bottle was determined by the serial dilution method. Meat infusion broth with 1% neopeptone at pH 7.4 was used as diluent. A strain of Lancefield Group A beta hemolytic streptococci was chosen at random as the test organism. Each tube of diluted penicillin was seeded with a drop of an 18-hour broth culture containing about 30 million organisms.

Cultures having effective titers in a dilution of 320 or over were pooled. The pH was adjusted to 6 and 500 ml were poured into a 700 ml distilling flask through cheesecloth. The flask was partly immersed in a water bath the temperature of which was maintained at 60°C. The substrate containing penicillin was evaporated under reduced pressure. The process is illustrated by the accompanying diagram (Fig. 1). A vacuum of about 0.001 mm of mercury was created by a running water pump or the Cenco-Hyvac pump. In 3 hours, the volume of 500 ml could be reduced to 20 ml. The pH of the concentrated stuff was readjusted to 6 and it was sterilized by filtration. While the volume of the crude penicillin was reduced, the titer of the active principle increased manyfold.

The concentrated penicillin was kept in celluloid tubes. Each lot was divided into two portions; one was placed in a dry-ice

1 Fleming, A., *Brit. J. Exp. Path.*, 1929, **10**, 226.

2 Hobby, Gladys, and Myer, Karl, PROC. SOC. EXP. BIOL. AND MED., 1942, **50**, 227.