

marked a tachycardia and had no effect on alveolar CO₂ tension.⁷

Summary. Smoking one cigaret according to a standardized exaggerated technic, lowers alveolar CO₂ for from 10 to 30 minutes, and increases blood pressure and heart rate for from 30 to 60 minutes in both smokers and relative non-smokers. Nicotine-free ciga­rets produce very much less effect.

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Effects of Transplanted Blastemas on Amputated Nerveless Limbs of Urodele Larvae.

OSCAR E. SCHOTTÉ, ELMER G. BUTLER AND ROBERT T. HOOD.
(Introduced by O. C. Glaser.)

From the Departments of Biology of Amherst College and of Princeton University.

Recent histological studies on early stages of Urodele regeneration have revealed that one of the primary consequences of limb amputation is the initiation of dedifferentiation at the amputation level. (Butler,^{1, 2} Puckett,³ and Thornton.^{4, 5}) It has been proposed by these authors that dedifferentiated cells contribute to the establishment of the regeneration blastema. The appearance of the blastema coincides with the cessation of dedifferentiation and marks the beginning of the regenerative processes.

Experimental evidence thus far presented indicates that an equilibrium of cellular interaction underlies regenerative activity. In particular, it has been suggested by Butler and Puckett⁶ that the blastema is important, not only in the establishment of a regenerate, but also in maintaining a balance between early dedifferentiation and later differentiation. When, by means of either X rays or ultraviolet light, regeneration is prevented, an active blastema never forms. Under such a condition the limb undergoes excessive dedifferentiation. Similar results have recently been obtained by Schotté and

⁷ Main, R. J., *Proc. Soc. Exp. Biol. and Med.*, 1940, **45**, 776.

¹ Butler, E. G., *J. Exp. Zool.*, 1933, **65**, 271.

² Butler, E. G., *Anat. Rec.*, 1935, **62**, 295.

³ Puckett, W. O., *J. Morph.*, 1936, **59**, 173.

⁴ Thornton, C. S., *J. Morph.*, 1938a, **62**, 17.

⁵ Thornton, C. S., *J. Morph.*, 1938b, **62**, 219.

⁶ Butler, E. G., and Puckett, W. O., *J. Exp. Zool.*, 1940, **84**, 223.

Butler^{7, 8} in preventing regeneration by denervation of limbs. Since no blastema ever forms on a completely nerveless limb, Butler and Schotté⁹ have drawn the conclusion that the excessive dedifferentiation in such limbs is a consequence of the absence of a blastema. It is of particular significance, however, that, if a blastema has been established before the time of radiation or of denervation, although regeneration is prevented, excessive dedifferentiation of structures does not occur. But, up to the present time, no experimental evidence has been available as to whether a transplanted active blastema can exert an influence on dedifferentiation.

The purpose of the present investigation, therefore, has been to transplant blastemas of various ages to nerveless limbs, which ordinarily would undergo extreme dedifferentiation, as demonstrated by Schotté and Butler.^{7, 8} A limb of a host larva was denervated by resection of the brachial plexus (left limb, larva C, Fig. 1); soon after denervation the limb was amputated, either through the metacarpals or the forearm region. To the cut surface of amputation was transplanted a regeneration blastema, secured from a previously amputated normal limb (right limb, larva A, Fig. 1).

1. *Transplantation of Blastemas Showing Early Digit Differentiation.* Approximately one hundred transplantations of blastemas showing digit formation have been carried out successfully. The effect of such transplanted blastemas consists essentially in retardation of the rate of limb regression, but not in the cessation of dedifferentiation. The blastema itself was affected by the dedifferentiation of the host limb and eventually was resorbed.

From these experiments it can be concluded that relatively old blastemas, which possess an appreciable amount of differentiation, do not exert an effective regulatory action on dedifferentiation of a nerveless host limb.

2. *Transplantation of Young Blastemas Showing no Evidence of Differentiation.* Results differing markedly from the above were obtained with young blastema transplants. In those cases in which a young transplanted blastema remained squarely in place on the amputation surface of the host limb, little or no regression of limb structures took place. Several cases have been followed for as long as two months, with no appreciable reduction in limb size and no indication of dedifferentiation of skeletal structures (left limb, larva D, Fig. 1). Histological examination of limbs amputated at the

⁷ Schotté, O. E., and Butler, E. G., *Science*, 1941, **93**, 439.

⁸ Schotté, O. E., and Butler, E. G., *J. Exp. Zool.*, 1941, **87**, 279.

⁹ Butler, E. G., and Schotté, O. E., *J. Exp. Zool.*, 1941, **88**, 307.

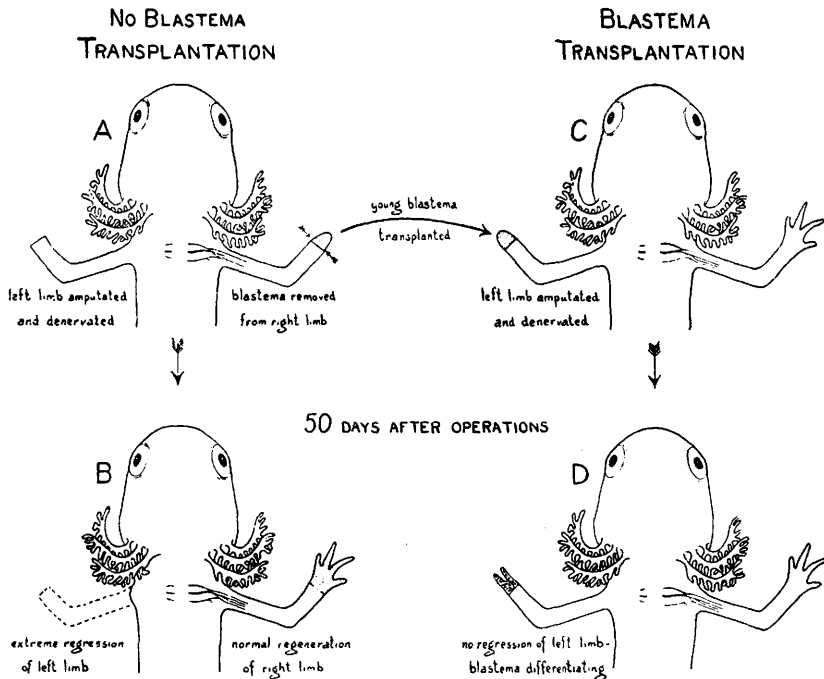


FIG. 1.

Schematic representation of the described experiments. The larva at the left (A and B) demonstrates the effects of amputation on a nerveless limb (according to Schotté and Butler⁸). The larva at the right (C and D) shows that, if a young blastema is transplanted to a limb which is then kept nerveless for a period of approximately a month and a half, no appreciable reduction occurs. While the transplanted blastema acts as a check to the dedifferentiation of the nerveless limbs, morphogenesis of the blastema, although generally defective, continues.

level of the carpals, to which young blastemas were transplanted, reveals no evidence of dedifferentiation at the end of 2 months. During this period control denervated limbs, amputated at the same level, underwent complete reduction proximally as far as the shoulder (left limb, larva B, Fig. 1).

Conclusions. These results provide new experimental evidence toward a further understanding of the rôle which the blastema plays in regeneration. It has been previously established (Schotté and Butler,^{7, 8} Butler and Schotté⁹) that in normal regeneration cessation of dedifferentiation of formed structures of the limb coincides with the appearance of a blastema. It has been demonstrated, furthermore, that when the formation of a blastema is prevented by denervation, dedifferentiation is unchecked and proceeds to extreme ends. In the experiments here reported it is shown that this extreme dedifferentiation in an amputated nerveless limb is prevented by the transplantation of a young blastema.

The presence of a blastema, therefore, is essential for the cessation of dedifferentiation and hence for the establishment of an equilibrium in the cellular interactions which occur during the initial phases of regeneration. This is true, however, only for young undifferentiated blastemas. When a transplanted blastema itself shows unmistakable signs of differentiation, it loses the capacity to control the dedifferentiation of a nerveless amputated limb.

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**Lung Volume and Pulmonary Dynamics in Raynaud's Disease.
Effect of Exposure to Cold.**

M. D. ALTSCHULE, H. LINENTHAL AND N. ZAMCHECK.

From the Medical Service and Medical Research Laboratories, Beth Israel Hospital, and the Department of Medicine, Harvard Medical School, Boston.

The occurrence of diffuse pulmonary fibrosis in patients with Raynaud's disease and scleroderma, though rare, has been noted in several instances.^{1, 2} The question, therefore, arises as to whether the lung volume and pulmonary dynamics are abnormal in patients with Raynaud's disease and, more particularly, whether exposure to cold affects their pulmonary function. Three patients have been studied; all were females and ranged in age from 15 to 39 years. One of them (Case 3) had cough, dyspnea and X-ray evidence of diffuse pulmonary fibrosis of the type associated with Raynaud's disease.² No such findings were present in the other two. All studies were made by the method of Christie,³ with the patients in the recumbent position. Measurements were made before and again after the patients' legs and at least one hand had been exposed to ice.

Observations. The lung volume and pulmonary dynamics in the 2 patients (Cases 1 and 2) with no clinical or X-ray evidence of pulmonary fibrosis were within normal limits. Case 3, in whom evidences of pulmonary fibrosis were present, exhibited a striking decrease in total pulmonary capacity, vital capacity and complemental air, with a lesser decrease in reserve air; the residual and functional residual airs were within normal limits. The respiratory rate and minute volume were increased.

¹ Matsui, S., *Mitt. a. d. med. Fakult. d. k. Univ. zu Tokyo*, 1924, **81**, 55.

² Linenthal, H., and Talkov, R., *New Eng. J. Med.*, 1941, **224**, 682.

³ Christie, R. V., *J. Clin. Invest.*, 1932, **11**, 1099.