origin and insertion (Figure 1). In another group of 6 frogs, a portion of iliac nerve from the middle of the femur to the posterior tibio-fibular branches was removed carefully without injuring the major vascular supply, followed by the removal of gastrocnemius muscle.

Results and discussion. Two animals from each group were sacrificed regularly 1, 2 and 7 months after the operation. In the group where the nerve supply was kept intact, 1 month after ablation the muscle regenerate was seen as a small band, 8 mm in diameter, extending between the origin and insertion, but connections were not established by most of the muscle bands (Figure 2). 2 months after the operation, the muscle regenerate appeared extending from the origin to the insertion. The regenerated muscle was about half of the original size, 13 mm in diameter, but lacked the normal shape (Figure 3). Regeneration was complete by 7 months after the removal. Here the muscle was almost identical to the control muscle of the opposite limb, except for its slightly reduced size (Figure 4).

Where the nerve has been removed before cutting the muscle, only a healing of the cut ends occurred and no trace of regeneration was evident, even after 7 months of ablation.

What has been recorded by other workers relates mainly to regeneration after mincing and implanting muscle fragments. The present study reveals for the first time the spontaneous regenerative ability of the gastrocnemius muscle of frog and also the significance of the nerve in muscle regeneration process[11,12].

Zusammenfassung. Die spontane Neubildung des Gastrocnemiusmuskels und die Rolle des Nerven in der Muskelneubildung des Frosches *Rana tigrina* sind untersucht worden.

R. RAVINDRANATHAN and P. MURALIDHARAN[12]

Department of Zoology, University of Kerala, Kariavattom P.O., Trivandrum (S. India), 16 April 1973.

[1] S. M. Ross, Ann. N.Y. Acad. Sci. *49*, 818 (1948).
[2] M. SINGER, *Regeneration of Vertebrates* (Ed. C. S. THORNTON; Chicago, Univ. Press, Chicago 1959), p. 59.
[3] B. M. CARLSON, J. Morph. *125*, 447 (1968).
[4] C. S. THORONTON, Am. Zoolog. *10*, 113 (1970).
[5] C. S. THORONTON, J. Morph. *62*, 17 (1938).
[6] E. D. HAY, Devel. Biol. *1*, 555 (1959).
[7] A. J. SCHMIDT, J. exp. Zool. *149*, 171 (1962).
[8] A. J. SCHMIDT, Anat. Rec. *155*, 65 (1966).
[9] A. N. STUDITSKY, Izdatel. Akad. Nauk. SSSR, Moscow, *1959*, 338.
[10] B. M. CARLSON, Anat. Rec. *166*, 423 (1970).
[11] O. E. SCHOTTE', Rev. Suisse. Zool. *33*, 1 (1926).
[12] M. SINGER, Q. Rev. Biol. *27*, 169 (1952).
[13] We express our gratitude to Dr. K. K. NAYAR and Dr. V. K. K. PRABHU for going through the manuscript and offering valuable suggestions. The work was supported financially with a grant from ICAR.

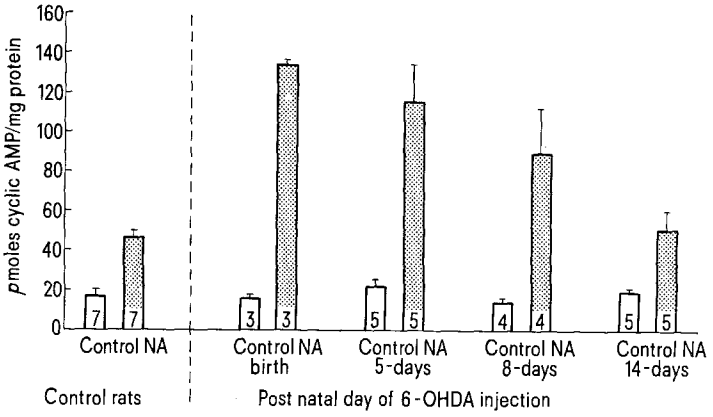
The Cyclic AMP Response to Noradrenalin in Young Adult Rat Brain Following Post-Natal Injections of 6-Hydroxydopamine[1]

Catecholamines readily stimulate the formation of cyclic 3′,5′-adenosine monophosphate (cyclic AMP) in incubated tissue slices of rat brain[2–4]. Moreover, an involvement of cyclic AMP in adrenergic transmission processes has been described[5]. Intraventricular injections of 6-hydroxydopamine (6-OHDA) which produce a selective destruction of catecholamine nerve endings in the rat brain[6–11], subsequently lead to an enhanced stimulation of cyclic AMP by noradrenalin (NA) in vitro[3,4]. Furthermore, during postnatal development of the brain prior to the appearance of monoamine containing nerve endings[9,12,13] the adenyl cyclase receptor that is responsive to these neurohormones exists in a similar hyperactive condition[14]. Recently it was reported that injections of 6-OHDA into rodents at birth resulted in behavioral changes, weight loss and deficits in brain NA content when the animals reached adulthood[9,15–17]. In the present study 6-OHDA was injected intracranially into rats at various times after birth in order to assess the effect of destruction of adrenergic neurons on the subsequent NA-induced stimulation of cyclic AMP in incubated tissue slices of cerebral cortex from young adult animals.

Methods. The experiments were carried out using Sprague Dawley-Holtzman rats. Littermates were injected intracranially with either a control solution (0.1% ascorbic acid) or 6-OHDA (250 µg in 0.1% ascorbate in 10 µl) at various times after birth and were maintained with their respective mothers. At 35 days postpartum the animals were sacrificed and the cerebral cortices removed, sliced and preincubated for 30 min in Krebs-Ringer bicarbonate buffer. The buffer was changed and after 15 min further incubation, NA (10^{-5} *M*) was added. 6 min later the samples were homogenized and cyclic AMP was isolated and determined by methods described previously[2–4]. Cyclic AMP is expressed as picomoles per mg sample protein.

Results. The results of the present experiments are depicted in the Figure. In the presence of NA cyclic AMP levels were consistently elevated 2–3-fold in the

[1] This investigation was supported by a research grant awarded by the National Institute of Mental Health No. MH 21584.
[2] G. C. PALMER, F. SULSER and G. A. ROBISON, Neuropharmacology *12*, 327 (1973).
[3] G. C. PALMER, Neuropharmacology *11*, 145 (1972).
[4] A. KALISKER, C. O. RUTLEDGE and J. P. PERKINS, Molec. Pharmac. *9*, 619 (1973).
[5] G. R. SIGGINS, E. F. BATTENBERG, B. J. HOFFER, F. E. BLOOM and A. L. STEINER, Science *179*, 585 (1973).
[6] F. E. BLOOM, S. ALGERI, A. GROPPETTI, A. REVUELTA and E. COSTA, Science *166*, 1284 (1969).
[7] U. UNGERSTEDT, Eur. J. Pharmac. *5*, 107 (1968).
[8] N. J. URETSKY and L. L. IVERSEN, J. Neurochem. *17*, 269 (1970).
[9] G. R. BREESE and T. D. TRAYLOR, Br. J. Pharmac. *44*, 210 (1972).
[10] D. M. JACOBOWITZ, in *Frontiers in Catecholamine Research*, III International Catecholamine Symposium, 1973, in press.
[11] K. NAKAMURA and H. THOENEN, Psychopharmacologia *24*, 359 (1972).
[12] H. C. AGRAWAL, S. N. GLISSON and W. A. HIMWICH, Biochim. biophys. Acta *130*, 511 (1966).
[13] G. K. AGHAJANIAN and F. E. BLOOM, Brain Res. *6*, 716 (1967).
[14] M. J. SCHMIDT, G. C. PALMER, W.-D. DETTBARN and G. A. ROBISON, Devel. Psychobiol. *3*, 53 (1970).
[15] B. SINGH and J. DE CHAMPLAIN, Brain Res. *48*, 432 (1972).
[16] O. CASTREN, M. AIRAKSINEN and S. SAARIKOSKI, Experientia *29*, 576 (1973).
[17] I. CREESE and S. D. IVERSEN, Brain Res. *55*, 369 (1972).



Rats were injected intracranially at various times after birth with either a control solution or 6-OHDA. At 35 days post-partum the animals were sacrificed and the cerebral corties removed, sliced and preincubated at 37 °C for 30 min in Krebs-Ringer bicarbonate buffer. The buffer was changed and the incubations were continued for 15 min at which time NA ($10^{-5}M$) was added. 6 min later the samples were homogenized, cyclic AMP was isolated, measured and expressed as picomoles per mg sample protein. The values in the Figure represent the mean ± SEM and the number of separate experiments are noted at the base of the individual bar graphs.

cerebral cortex of 35-day-old control rats. The cyclic AMP response to NA was 6–9-fold in the animals injected with 6-OHDA at either birth, 5 or 8 days postpartum. No enhanced accumulation of cyclic AMP in response to NA was seen in the incubated cerebral cortices of the animals injected with 6-OHDA at 14 days after birth. In these and earlier studies[3,4] basal levels of the cyclic nucleotide were not affected to any appreciable extent following the intracranial injections of 6-OHDA. In support of previous findings[3,4,16] the 6-OHDA injected rats that demonstrated a 'supersensitive' cyclic AMP response to NA were considerably smaller in size than control injected littermates and did not demonstrate any post-decapitation muscular activity. Moreover, these drug treated animals when handled manifested an increased irritability that was characterized by a fierce attacking behavior similar to that initially reported by PALMER[3] and subsequently described in detail by NAKAMURA and THOENEN[11].

Discussion. From the present experiments it is readily evident that intracranial injections of 6-OHDA at either birth or shortly thereafter lead to a consequent alteration of the receptor moiety of adenyl cyclase in the cerebral cortex when the animals reached the young adult stage. This hyperactive condition of the receptor might reflect an involvement of adenyl cyclase in the underlying molecular mechanisms associated with the phenomenon of adrenergic denervation supersensitivity. Similar investigations using adult rats have described a hyperactive adenyl cyclase following chemical destruction or depletion of adrenergic nerve endings[2–4]. Likewise, additional studies have reported that destruction of catecholamine containing nerve endings leads to supersensitive responses of amphetamine on dopamine receptors[17] and NA actions on isolated atrial and duodenal preparations[18,19].

6-hydroxydopamine appears to selectively destroy adrenergic nerve endings[6,8–11], however, a direct neurotoxic effect on cell bodies has been described[7]. Whether or not these actions of 6-OHDA are permanent in adult or developing animals is subject to considerable debate[9,10,15,17,18,20]. In the rat brain the development of central monoamine containing neurons and respective synaptic connections comes about after birth[9,12,13]. Perhaps in the present study the early injections of 6-OHDA (birth, 5 and 8 days postpartum) destroyed the fetal adrenergic neurons before the capacity to regenerate had appeared. In this case the synaptic inputs that would normally exert a controlling influence on the receptor sites remained absent and the immature hyperactive enzyme persisted throughout development. Conversely, the animals injected at 14 days postpartum may have reached a state of maturation such as to either develop compensatory mechanisms or regenerate sufficient adrenergic nerve endings that would ultimately exert a normal modulating action on the post synaptic adenyl cyclase receptor. These preliminary observations on the development of the adenyl cyclase receptor should provide a framework for further investigation.

Zusammenfassung. Nachweis, dass neugeborene Ratten (bis zum 8. Tag), die intracerebral mit 6-OH-Dopamin behandelt wurden, am 35. Lebenstag eine Überempfindlichkeit gegenüber der Adenylzyklase-stimulierenden Wirkung von Noradrenalin aufweisen.

G. C. PALMER and H. R. SCOTT

Department of Pharmacology, University of New Mexico School of Medicine, 915 Stanford Drive N.E., Albuquerque (New Mexico 87131, USA), 13 November 1973.

[18] J. DE CHAMPLAIN and R. NADEAU, Fedn. Proc. *30*, 877 (1971).
[19] J. F. GIUDICELLI, Experientia *27*, 1194 (1971).
[20] G. M. LEW and W. B. QUAY, Res. Commun. Chem. Path. Pharmac. *2*, 807 (1971).

Differences in Brain and Body Weights of Mice Caused by Differential Housing[1]

It is becoming increasingly evident that significant differences in the gross and microscopic morphologies of the brains of rodents can be produced by simply changing their housing conditions[2–13]. The changes which differential housing causes in cerebral gross morphology, though slight, can be correlated with more pronounced changes in gross behavior[8,13,14–19], in brain chemistry and metabolism[8,13,15,19,20–26], and in the pharmacological effects