ing agent, blocks the agonistic effect of norepinephrine on the β receptor thus allowing a maximal melanin granule aggregating response resulting from an uninhibited α adrenergic receptor stimulation.

Discussion and conclusions. The observations made in this study indicate that the thermal polymer: arginyl, glutamyl, glycyl, histidyl, phenylalanyltryptophan not only stimulates melanin granule dispersion within melanophores but stimulates iridophore contraction as well. The combined effect of this material on both the iridophores and melanophores of *Rana pipiens* is responsible for the reflectance changes observed. While the decrease in reflectance of frog skins was admittedly weak it was definite and consistent and apparently of a slightly greater magnitude than that observed by Fox and Wang[4] in an in vivo method using the hypophysectomized frog[12].

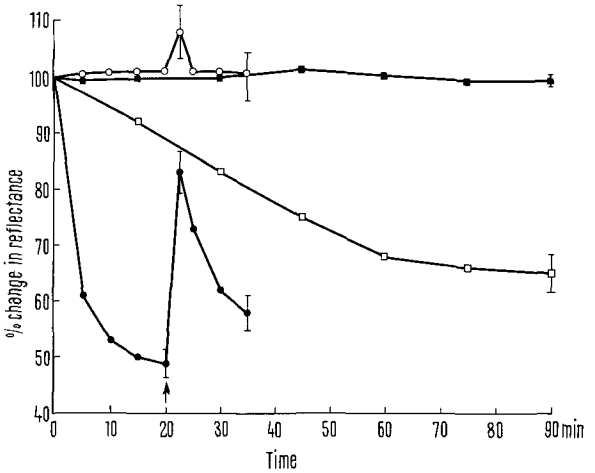


Fig. 4. Comparative in vitro response of *Rana pipiens* skins and *Anolis carolinensis* skins to a thermal polymer. One group of *Rana* (□) and one group of *Anolis* (●) skins were incubated in a 5×10^{-4} g/ml concentration of the peptide. One group of *Rana* (■) and one group of *Anolis* (○) skins were maintained as Ringer controls. At 20 min (arrow), norepinephrine ($10^{-5} M$) was added to both groups of *Anolis* skins. Results are means of the reflectance measurements from 8 skins per point on the graph.

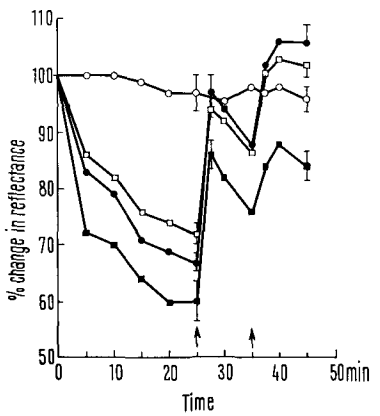


Fig. 5. Three groups of *Anolis* skins were darkened by a thermal polymer at one of the following concentrations: 10^{-5} (□), 5×10^{-5} (●), or 10^{-4} (■) g/ml. One group (○) of skins was maintained as a Ringer control. At 25 min (arrow), norepinephrine ($10^{-5} M$) was added to the 3 groups of darkened skins and this was followed at 40 min (arrow) by the addition of dichloroisoproterenol ($10^{-4} M$) to these same skins. Results are means of the reflectance measurements from 8 skins per point on the graph.

The experiments in which *Anolis* skins were used are of particular importance because the response of reptilian melanophores to the polymer is similar to their normal response to MSH. This response can be reversed by a normal mechanism of catecholamine stimulation involving α adrenergic receptor stimulation. This is the first demonstration that melanin granule dispersion in response to an MSH-like peptide can be reversed by another hormone. Apparently the mechanism of catecholamine antagonism of MSH which results in melanin granule aggregation is operative through the same small portion of the parent MSH molecule as is responsible for causing dispersion.

Of particular significance in these experiments is the demonstration that the chromatophores of *Rana* and *Anolis* respond differentially to the hexatonic polymer. Fox and Wang[4] point out that it is not merely the presence of a 6-component polymer which accounts for the chromatophore activity, but rather, there is a biological specificity attributable to the polymer used. It may be that *Anolis* melanophores respond more strongly to the polymer than *Rana* because the amino acid sequence of the thermal polymer resembles more closely that of the naturally occurring intermedin of *Anolis* than *Rana*. This is not unreasonable in view of the fact that it has been shown that *Anolis* has an intermedin which differs from that of mammals[12,13].

Zusammenfassung. Der Einfluss der Hitzepolymerisate Arginin, Glutaminsäure, Histidin, Phenylalanin und Tryptophan auf die Chromatophoren wurde geprüft. Die Reptilienhaut (*Anolis carolinensis*) zeigte auf Grund von photometrischen Reflexionsmessungen eine stärkere MSH-Reaktion gegenüber einem synthetischen kleinen Peptid als die Amphibienhaut (*Rana pipiens*). Die hautverdunkelnde Wirkung kann durch Noradrenalin und einen Blocker wieder wieder aufgehoben werden.

J. T. Bagnara and M. E. Hadley

Department of Biological Sciences,
University of Arizona,
Tucson (Arizona 85721, USA), 13 August 1969.

[12] A. C. J. Burgers, Abst., 1st Intern. Congr. Endocrin., Copenhagen, *165*, 329 (1960).

[13] This study was supported by grant No. GB-8347 from the National Science Foundation.

Reversible Lenticular Opacities Induced in Rats by Emotional Stress

Adrenaline[1] or morphine type drugs[2] which release adrenaline from the adrenal medulla[3] have been found to produce acute reversible cataracts in rodents in conjunction with lack of lid reflex movements and exophthalmos. These lens opacities can be prevented by any drug which prevents the eyes being kept wide open[4] or by the closure of the eye[5]. This observation gave support to the idea that dehydration is the major stimulus of this type of

cataract formation and proptosis is responsible for the increased evaporation through the cornea[4,6]. However, the observations reported here suggest that a more complex pharmacological action may be involved in the process.

In the course of experiments when we studied the effect of different catecholamines on the lens of 45- to 55-day-old male albino rats of Porton-Wistar strain 2 h after the s.c. administration of 150 mg/kg pargyline hydrochloride (Abbott Laboratories Ltd., Queensborough, Kent), it has been found that after repeated examination 2 of 10 rats showed signs of transient sucapsular opacities without the administration of any catecholamine. To increase pupillary size 0.5% w/v cyclopentolate (Ward, Blenkinsop & Co. Ltd., London) was dropped into the eyes. The animals were observed under the slit lamp (Zeiss Photo Slit Lamp, Oberkochen, Germany) while fixed manually, and it was thought that this treatment represented a stressful situation which via catecholamine release activated the process responsible for the formation of acute lenticular opacities in some of the more sensitive animals. When the experiments were repeated with 10 animals not treated with pargyline and 10 other animals treated with pargyline but anaesthetized with 0.84 mg/kg Nembutal 20 min prior to observation, none of them showed any signs of lenticular opacities. However, when 20 rats pretreated with pargyline were placed one by one in a sound-proof box (size $18 \times 17 \times 13$ inches) in which an externally controlled door-bell and flashing light operated for 20 sec with 13 sec breaks over a period of 2 min, 16 rats developed dense subcapsular opacities 20–40 min after the exposure. The opacities disappeared 40–60 min later. In a group of 8 rats that has previously been accidentally subjected to stress situation of continuous light for at least 2 weeks due to switch failure in the animal house, only 2 developed slight opacities.

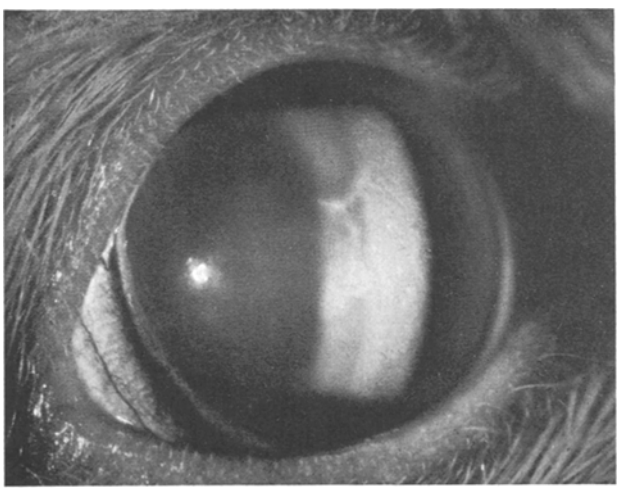
The significant increase in the frequency and density of lenticular opacities by pretreatment with an amine-oxidase inhibitor and the effect of a long-lasting stress situation prior to the experimental emotional stress, underlines the importance of the release of catecholamines in the development of acute lenticular opacities induced by emotional stress. However, the lenticular opacities caused by adrenaline and by emotional stress are not comparable. In rats the lethal and cataract-producing doses of adrenaline are so near to each other that 2.5–5.0 mg/kg adrenaline killed 63% of the animals within 1 h and only 6.5% developed cataracts[6]. Contrary to this, after emotional stress the animals looked quite normal, without any sign of adrenaline intoxication, had no exophthalmos and retained lid reflex, and in spite of this 80% of the animals developed cataracts. It might be that other catecholamines released had a synergetic effect on adrenaline, but it is more likely that catecholamines released from the brain and moreover from iris and ciliary body are in a better position to activate the process than the i.p. injected adrenaline. This latter possibility is supported by the fact that levorphanol, a morphine derivative, produced opacities in a much lower dose when it was given intracerebrally than i.p.[8].

The effect of iridial noradrenaline on the aqueous flow and occular pressure has been well established[8,9]. It might be that noradrenaline released in the eye by the emotional stress resulted in a change both in the outflow and composition of the aqueous humour. This change by increasing the surface tension of the aqueous layer in contact with the oily layer disrupted the latter. Though the closure of the eye prevented the development of lenticular opacities, it seems unlikely that lid retraction resulting in increased evaporation through the cornea is the sole factor responsible for the development of acute reversible lenticular opacities. Opacities appeared 20 min after stress and rats usually blink only once or twice in every 20 min so the lack of blinking could affect the oily layer and the lens after a much longer time. Furthermore, in our experiments animals developed lenticular opacities with no signs of exophthalmos and it has been reported that lenticular opacities induced by blowing air across the cornea disappeared even while the stimulus persisted[10].

Zusammenfassung. Ratten (20 Tiere, 45 Tage alt) erhielten 150 mg/kg paraglynes Hydrochlorid s.c. injiziert. Akustischer Schock (2 h später) ergab bei 16 von 20 Tieren eine reversible subkapsuläre Linsentrübung.

F. O. MUELLER and L. MAGOS[11]

Toxicology Research Unit,
Medical Research Council Laboratories,
Carshalton (Surrey, England), 25 July 1969.



Slit-lamp photograph of the left eye of a rat 20 min after emotional stress. 120 min before stress the animal was given s.c. 150 mg/kg pargyline hydrochloride. Note the irregular cornea surface (right light-beam) and the anterior cortical cataract mainly below the noticeable Y-shaped anterior suture line (left light-beam).

[1] C. TUM SUDEN, Am. J. Physiol. *130*, 543 (1940).
[2] M. WEINSTOCK, H. C. STEWART and K. R. BUTTERWORTH, Nature *182*, 1519 (1958).
[3] L. M. GUNNE, Acta physiol. scand. *58*, suppl. 204, 5 (1963).
[4] M. WEINSTOCK, Naunyn-Schmiedeberg's Arch. exp. Path. Pharmak. *259*, 201 (1968).
[5] F. T. FRAUENFELDER and R. P. BURNS, Arch. Opthal. *76*, 599 (1966).
[6] C. TUM SUDEN and L. C. WYMAN, Endocrinology *27*, 628 (1940).
[7] A. SMITH, M. KARMIN and J. GAVITT, J. Pharm. Pharmac. *18*, 545 (1966).
[8] K. E. EAKINS and H. M. T. EAKINS, J. Pharmac. exp. Ther. *144*, 60 (1964).
[9] M. J. ROSSER and M. L. SEARS, J. Pharmac. exp. Ther. *164*, 280 (1968).
[10] F. T. FRAUENFELDER and R. P. BURNS, Proc. Soc. exp. Biol. Med. *110*, 72 (1962).
[11] Acknowledgement. The authors wish to thank Mr. R. D. LOCK for technical assistance.