

taining approximately 25 growth hormone units per mg (body weight test in hypophysectomized rats),⁷ but less than 1% of any of the other known pituitary hormones. It will be noted that in the case of the growth hormone, markedly larger effects were secured, increasing with increasing dosage. This was not the case with any of the other hormones here employed.

Summary. 1. Purified anterior hypophyseal hormones other than the growth hormone were

⁶ Marx, W., Simpson, M. E., and Evans, H. M., *J. Biol. Chem.*, 1943, **147**, 77.

⁷ Marx, W., Simpson, M. E., and Evans, H. M., *Endocrinology*, 1942, **30**, 1.

administered to hypophysectomized rats for a period of 4 days and measurements made of the width of the uncalcified portion of the proximal epiphyseal cartilage of the tibia. 2. While adrenotropic hormone slightly decreased the cartilage width, thyroxin and the thyrotropic as well as the mammatropic hormones caused slight enlargement of the epiphyseal cartilage. The effect was smaller, however, than the response caused by a single growth hormone unit, at all dose levels tested, and the effect did not show gradation with dosage which is a characteristic of the growth substance.

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A Comparison of Regeneration and Respiration Rates of Tubularia.*

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Barth¹ showed that oxygen can be made the determining factor in the polarity and rate of regeneration of Tubularia stems. In an attempt to find the metabolic basis of regeneration, Barth² investigated the relation between oxygen consumption rates and regeneration rates, and found a general parallelism between the two at various oxygen tensions. In the same study a comparison of the oxygen consumption rates of regenerating versus resting stems showed comparatively little difference between the two groups. This important finding indicated that while the process of regeneration depended on some portion of the aerobic metabolism, a relatively small part of the overall respiration was involved. Moog and Spiegelman³ were able to show with the

use of various respiratory poisons that complete inhibition of regeneration could be obtained without any parallel effect on respiration that was measurable.

These findings imply that the stimulatory effect on regeneration of increased oxygen tensions is a non-specific one and can be interpreted as a general influence on all oxygen consuming systems.

Goldin⁴ found that at oxygen tensions at which normal regeneration rates might be expected, complete inhibition could be obtained by lowering the hydrogen ion concentration sufficiently. Therefore it was of importance to determine whether the pH effect on regeneration was mediated through its effect on the overall oxygen consumption or whether it was of a more specific nature such as was found with some of the respiratory inhibitors.

Methods. Young, unbranched Tubularia stems of uniform diameter were selected from colonies gathered from the waters of Cape Cod

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¹ Barth, L. G., *Physiol. Zool.*, 1938, **11**, 179.

² Barth, L. G., *Biol. Bull.*, 1940, **78**, 366.

³ Moog, F., and Spiegelman, S., *Proc. Soc. Exp. Biol. and Med.*, 1942, **49**, 392.

⁴ Goldin, A., *Biol. Bull.*, 1942, **82**, 243.

Bay during the months of July and August. Stem segments, 6 mm in length, were cut from the region 5-11 mm proximal to the hydranth. The stem segments were suspended in filtered sea water, the oxygen tension and pH of which were adjusted by saturation with appropriate mixtures of oxygen, carbon dioxide, and nitrogen. The oxygen tension was kept constant at that of normal sea water, the pH being varied by means of the CO₂-nitrogen ratios.

Oxygen consumption values were obtained with Warburg manometers and vessels. All experiments were run at $19 \pm 0.1^\circ\text{C}$. The manometers were shaken at a rate of 90 oscillations per minute through a 5 cm arc. Each vessel was flushed with the gas mixture used to saturate the sea water in it. The bicarbonate buffer system required the use of the indirect method of Warburg.⁵ In this method 2 manometers are needed for each Q_{O₂} determination. The stems were suspended in 8 cc of sea water in one vessel and 3 cc in the other of a pair.

Wet weight determinations were made at the end of the run by means of a micro-balance after removal of excess moisture from the stem of the filter paper. Calculations are based on cu mm of O₂ consumed per mg of wet weight per hour. Hydrogen ion concentrations were determined with a glass electrode.

Results. Table I summarizes results of experiments covering a range of pH from 8.0 to 6.6. This pH range was found by Goldin⁶

TABLE I.

Variation of Respiration Rates with pH.

Rate of O₂ consumption in cmm/wet weight/hour of controls (pH 8.0) was 5.3 in Exp. 1, and 7.3 in Exp. 2. Stems were examined at end of run for signs of regeneration. + signifies regenerative activity; — signifies complete absence.

	pH	% normal respiration	• Regeneration
Exp. 1	8.0	100	+
	6.8	55	—
	6.6	27	—
" 2	8.0	100	+
	7.4	115	+
	6.7	46	—
	6.6	37	—

⁵ Warburg, O., *Biochem. Z.*, 1924, **152**, 51.

⁶ Goldin, A., *Biol. Bull.*, 1942, **82**, 340.

TABLE II.

Variation of Regeneration Rates with pH. Each value is the average of measurements on 30 stems. The *t* (hours) is the time to constriction. *L* is length of primordium at constriction.

pH	<i>L</i> in micra	<i>L/t</i>	1/ <i>t</i>	% normal <i>L/t</i>
8.07	1054	31.7	0.302	100
7.07	848	18.3	0.216	58
6.82	699	9.3	0.141	31
6.63	29	0.3	0.110	1

to include all degrees from no effect on regeneration, to complete inhibition. A determination of regeneration rates on the stems in the Warburg vessels could not be made since this would have involved opening the vessels and removing the stems for measurement during a respiratory experiment. Table II, however, gives some representative data of the variation of regeneration rate with pH. Regeneration rate is calculated as per cent of normal at pH 8.1. The data were collected in the usual fashion (Goldin, *loc. cit.*) at the same time that the respiration experiments were made, so that a comparison is possible.

Discussion. Table I shows that lowering the pH of the sea water medium is accompanied by a decrease in the respiratory rate. A comparison with the data in Table II reveals that within the same pH range a fall also occurs in the rate of regeneration.

It is evident that a parallelism, which is stronger at the lower pH values, exists between the effect of varied hydrogen ion concentrations on the two processes. Since a direct relation exists between respiration and regeneration (Barth, *loc. cit.*) it must be concluded that the greater part of the pH effect on regeneration is mediated through its effect on the rate of respiration.

It is probably that we have here, as in the case of cyanide (Moog and Spiegelman, *loc. cit.*) a non-specific inhibition of oxidative systems with a consequent depression of all activities dependent on them, among which is included the process of regeneration.

Summary. Lowering the hydrogen ion concentration produces a parallel depression of respiration and regeneration rates. It is concluded that the effect on regeneration is a non-specific one mediated through its effect on the enzyme systems involved in aerobic respiration.