ministration of both preparations of growth hormone, there was a retention of potassium, phosphorus, calcium, and sodium. The positive balances of the various substances studied was in part the result of a fall in their fecal excretion. There was a gain in body weight during both periods; the weight was maintained for 4 days after administration of the human material was stopped. There was a significant increase in urinary excretion of aldosterone during both periods of administration of growth hormone. This was most marked with the human growth hormone and was not accompanied by any alteration in urinary 17-hydroxy corticoid and 17-ketosteroid excretion. Impairment of the glucose tolerance curve was evident after 10 days of administration of the human growth hormone. No change in glucose tolerance was demonstrable on the fifth day of administration of monkey growth hormone.

The rather striking physiological effects obtained with human and monkey growth hormone in this study, if confirmed, are in contrast with the equivocal effects of the bovine preparations in man and lend support to the concept of species differences in the molecular structure of growth hormone. Recently Li (*15*) has reported on the difference in chemical and physical properties of bovine, monkey, and human growth hormone and has suggested that these various growth hormones possess a similar "core" with variations in the remaining part of the molecule which account for the species differences observed.

The effect of human and monkey growth hormone in causing an increase in aldosterone excretion in this patient is of interest in relation to previous investigations carried out with bovine growth hormone. Earlier studies by Venning *et al.* (*16*) with crude preparations of bovine growth hormone in healthy subjects suggested a stimulating effect on aldosterone excretion. When these experiments were repeated with highly purified preparations, Venning *et al.* (*17*) were unable to demonstrate any effect on the excretion of the hormone. Whether the present findings are the result of the specific effect of human and monkey growth hormone on aldosterone secretion or the result of some contaminant present in the preparations remains to be clarified.

The difference in the effect of the human and monkey growth hormone on the glucose tolerance curves carried out during its administration might either be due to differences in the length of administration of the hormone or to an actual species difference.

These data would suggest that the physiologically effective dose of human growth hormone prepared by the Raben technique is less than 10 mg per 24 hours, and further studies concerning this will be made as supply of the material permits.

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7 March 1957

Behavior, Sexual Dichromatism, and Species of Parrot Fishes

During a study of social behavior of parrot fishes (Scaridae) at the Bermuda Biological Station (*1*) in the summer of 1956, it became evident that many of the forms hitherto listed as species were of only one sex. Our first suspicion of this arose when we observed aggregations that included only two species. In each situation the aggregation included at least one brightly colored parrot fish among several-to-many inconspicuously colored ones. The sexual dimorphism is so pronounced that previous authors (*2*) have accorded species status to the male and female color phases.

Our methods of determining which male and female belonged together were (i) observation of the spawning aggregations and other associations of the color forms recognized, (ii) injection of testosterone into females of a certain color pattern, followed by observation of the resultant male color pattern, and (iii) comparison of morphologic characteristics. The list of synonomies and the reason for each action is given, in abbreviated form in subsequent paragraphs.

1) *Scarus gnathodus* (Poey) (female and immature) is a synonym of *Scarus vetula* (Block and Schneider) (male). This observation was first made by L. P. Schultz of the National Museum (Washington, D.C.), who could find no morphologic differences between the two currently recognized species except in the color pattern of adults. Because of the poor state of preservation of specimens available to him, he was unable to distinguish the sexes. These two forms are synonomized for the following reasons. All *S. vetula* examined were adult males; most adult-size *S. gnathodus* were females. The black-and-white colored *S. gnathodus* was the only form found in the immature stages. One adult of *S. vetula* was usually seen surrounded by from two to five adult *S. gnathodus* on the reefs. These were aggregations that were defended and led by the male *S. vetula*. Injections of testosterone caused adult females of *S. gnathodus* to transform into the bright blue and red pattern of *S. vetula*. Morphologically the two are similar with respect to scale numbers, body form, fin ray numbers, and so on.

2) *Scarus punctulatus*, Cuv. and Val. (male) is placed in the synonymy of *Scarus croicensis* Bloch (female and immature) on the basis of the following evidence. All *S. punctulatus* examined were males, and almost all adult-size *S. croicensis* were females. Only the black-and-white colored *S. croicensis* appeared in an immature stage. One or two adults of *S. punctulatus* were almost always in schools with several-to-many adult *S. croicensis*. Injections of testosterone into female *S. croicensis* transformed their black and white color into the bright blue-green and red color pattern of *S. punctulatus*. Morphologically the two are similar.

3) *Sparisoma distinctum* (Poey) (female and immature) is a synonym of *Sparisoma aurofrenatum* (Cuv. and Val.) (male). This statement is based on the following evidence. All *S. distinctum* examined were either adult females or immature; all *S. aurofrenatum* were adult males. Males of *S. aurofrenatum* were observed to spawn with females of *S. distinctum*. Adults of both, in the breeding season, were usually seen together in loose aggregations. Except for the more intricate color pattern of red and yellow, *S. aurofrenatum* is morphologically similar to *S. distinctum*.

4) *Sparisoma abildgaardi* (Bloch) (female and immature) appears to be a

synonym of *Sparisoma viride* (Bonnaterre) (male). The evidence, although not as strong as in the examples already cited, seems to warrant synonomizing these two forms. All *S. viride* examined were males, and all *S. abildgaardi* examined were females. Only immature stages of the *S. abildgaardi* red color phase were found, and only large, mature *S. viride* in their bright green phase were observed. During the breeding season, the adult red females of *S. abildgaardi* and the adult green males of *S. viride* were usually seen together in loose aggregations. Individuals of *S. viride* frequently chased individuals of *S. abildgaardi* but did not chase other parrot fishes. Also, it was noted by Longley (*2*) that *S. viride* has a spotted color phase similar to that of *S. abildgaardi*.

Brock and Yamaguchi (*3*) identified two species of Pacific Ocean parrot fishes as the male and female of each other. A similar finding has been described in the case of only one other species of parrot fish (*Sparisoma radians*), in the western North Atlantic Ocean (*2*).

These results further demonstrate the occurrence of sexual dimorphism in coral reef fishes and especially in the family Scaridae, which has been considered not to be sexually dimorphic, in general (*4, 5*).

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References and Notes

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19 February 1957

Synthesis and Pharmacology of the Octapeptide Angiotonin

Arterial hypertension of renal origin seems to depend, at least during its initiation, on the liberation of a pressor substance from the kidneys. In 1939 a substance with the theoretically required properties was discovered simultaneously by Page and Helmer (*1*) and Braun-Menendez, Fasciolo, Leloir, and Munoz (*2*). It resulted from the action of a proteolytic enzyme, renin, which was extracted from kidneys, on a substrate synthesized in the liver and contained in plasma.

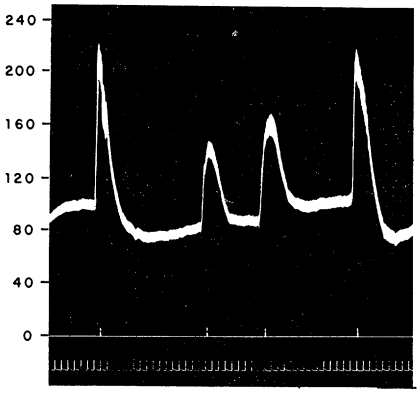

Fig. 1. Arterial pressure response in a dog under pentobarbital anesthesia and cardiovascular reactivity augmented with tetraethylammonium chloride. The marks on the abscissa indicate, left to right: (i) 5 μg of noradrenaline; (ii) natural angiotonin; (iii) synthetic angiotonin; (iv) 0.06 mg of serotonin.

More recently, Peart (*3*) isolated from the incubation mixture of renin (rabbit) and renin-substrate (beef) a pressor peptide containing ten amino acids (*4*) in the sequence L-asp, L-arg, L-val, L-tyr, L-val, L-his, L-pro, L-phe, L-his, L-leu (*5*). Skeggs *et al.* (*6*) obtained from the action of renin (hog) on its substrate (horse) a mixture of two vasoactive peptides, separable by countercurrent distribution, which they termed hypertensin I and II. They demonstrated that hypertensin I was converted into II by a chloride-activated enzyme occurring in plasma (*7*). Hypertensin I was shown to be a decapeptide with the sequence L-asp, L-arg, L-val, L-tyr, L-isoleu, L-his, L-pro, L-phe, L-his, L-leu; hypertension II was shown to be the corresponding octapeptide lacking the last two amino acids (*8*). Hypertensin I seemed to be inactive in the absence of converting enzyme (*7*).

In our work on the purification of angiotonin (hog renin, hog substrate), we were able to separate by countercurrent distribution an oxytocic-pressor principle from the dominantly pressor principle (*9*). The latter was transformed into the former by an enzyme present in whole, unhemolized blood, plasma, substrate fractions, hemolyzed red cells, and possibly urine. Distribution coefficients suggested a close relationship of the two principles with the two hypertensins of Skeggs *et al.* This was confirmed for the pressor principle, which we found to be identical with hypertensin I in amino acid composition and sequence.

On the basis of these data, it seemed very likely that the oxytocic-pressor principle was identical with hypertensin II. In order to determine the identity of these two principles, synthesis, based on the structure determined by Skeggs and coworkers for hypertensin II, was undertaken (*10*).

The four pure, crystalline dipeptides, cbz-β-Me-L-asp-NO_2-L-arg, cbz-L-val-L-tyr-Me, cbz-L-isoleu-L-his-Me, and L-pro-L-phe Me · HCl served as starting material. From the condensation of cbz-L-val-L-tyr-azide with L-isoleu-L-his-Me, a tetrapeptide was isolated. The carboxylic acid obtained from this tetrapeptide was condensed with L-pro-L-phe Me by the amide modification of the diethyl chlorophosphite method of Anderson and coworkers (*11*), to give the hexapeptide cbz-L-val-L-tyr-L-isoleu-L-his-L-pro-L-phe Me. The mixed anhydride formed from cbz-β-Me-L-asp-NO_2-L-arg-L-val-L-tyr-L-isoleu-L-his-L-pro-L-phe Me (mp, 182 to 185°C; $\alpha_D^{25} = -60.1$, $c = 1$ in methanol; $\alpha_D^{25} = -32.5$, $c = 1$ in dimethylformamide). After the removal of the protecting groups by hydrolysis and hydrogenolysis, a biologically active solution was obtained (20,000 units/mg of N). From this solution, a white powder was isolated containing a strongly pressor material and NaCl. After correction for NaCl, the activity was found to be 4000 units/mg of solid and by nitrogen determination as 22,000 units/mg of N (*9*). Pure natural angiotonin has an activity of 45,000 units/mg of N and 7800 units/mg of solid. Further purification of the synthetic angiotonin will be necessary before final comparison is justified. The material was also very active

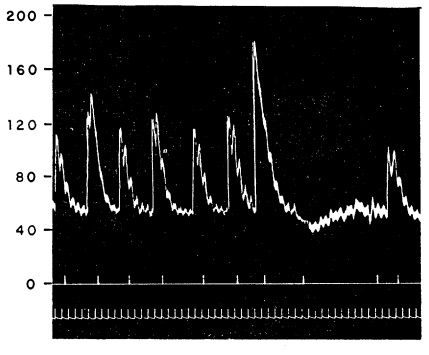

Fig. 2. Arterial pressure response in a pithed cat, comparing natural and synthetic angiotonin. The marks on the abscissa indicate, left to right: (i) 0.35 μg of solid synthetic angiotonin; (ii) 2 units of natural angiotonin; (iii) 0.41 μg of synthetic angiotonin; (iv) 1.6 units of natural angiotonin; (v) 0.41 μg of synthetic angiotonin; (vi) 0.47 μg of synthetic angiotonin; (vii) 5 μg of noradrenaline; (viii) 4 mg of benzodioxane per kilogram; (ix) 5 μg of noradrenaline; (x) 0.47 μg of synthetic angiotonin.