tioned secretions based on food and on acid, as well as the concomitant overt behavior, in radically different ways. The effect of variation in intensity of the conditioned stimulus and of constant stimuli other than hunger was compared with the effect of variations in the hunger drive and was also found to be quite different. An analysis of the difference in the effect of change of hunger upon conditioned and unconditioned secretion is presented. The results are inconsistent with the particular assumptions of traditional conditioning theory which the experiments were designed to test. It is felt that they bear significantly upon the interrelation of the general problems of motivation and learning.

Factorial analysis of learning dynamics in animals: ROBERT J. WHERRY (introduced by Walter R. Miles). The number of forces which have been hypothecated to explain animal learning are extremely numerous, one writer having listed over ninety factors. Certain attempts at correlation analysis, however, have endeavored to show that all errors during learning are in large part due to a single factor present *before* learning begins. Other attempts have tried to prove that another single factor developed *during* learning controls its course. In such studies, the correlations used were between the errors on the first run and the total error scores upon all later runs as a unit. Furthermore, such studies have assumed that the factor or factors involved are of a static nature. In the present study, factorial analyses were made of the learning by animals of maze and discrimination problems. These analyses involved correlations of scores on individual trials and alleys, thus bringing out the rise and wane of different factors. Further research indicated by these preliminary analyses is suggested, and its field of probable usefulness is specified.

Recovery sequence after anesthetization. II. Cyclopropane and nitrous oxide: ALBERT C. CORNSWEET (introduced by C. L. Hull). This is a further study of the behavioral tendencies exhibited by albino rats upon anesthetization. A previous study with ether used as the anesthetizing agent was reported at the 1936 meeting of the American Association for the Advancement of Science. In the present study, cyclopropane and nitrous oxide were used. The animals were subjected to varying amounts of the anesthetics until the animals were in a complete state of quiescence. Upon removal from the anesthetization chamber, the animals were stimulated by means of a tweezers-aesthesiometer, and observations were made as to the temporal sequence of the behavioral patterning. In general, the animals portrayed a sequence of movements, in a fairly definite cephalo-caudad direction. Movements in the head region occurred before those of the caudal extremities. These results coincided with the findings of ether anesthetization. The only consistent difference between ether anesthetization and these gases was that the latter's effect upon recovery was more rapid and telescoped. Of especial importance, however, was the fact that the animals upon going under the anesthetic exhibited caudocephalad behavior movements, a sequence that was the reverse of that of recovery. This fact is contrary to medical theory of anesthetization on human subjects, where the order is, supposedly, cerebrum, spinal cord and medulla. No attempt is made to lay down any definite hypothesis, for much more experimentation remains to be done. Many investigators on pre- and post-natal behavior have tended to emphasize one part of the organism's anatomical, structural and psychological constituents more than another; at times slighting the fact that the animal is a totality, a functional whole, and more than automaton made up of discrete units. Explanations have been too reflexological, rather than in terms of an observable whole. Further work is planned, using other species of life and other types of anesthetics, and perhaps then will a comparative correlation and theory be presented.

SPECIAL ARTICLES

THE SECRETION OF IODINE BY THYROID GLANDS CULTIVATED IN THE LINDBERGH PUMP

THE form in which iodine is secreted by the thyroid is not known. The work of some investigators indicates that it is secreted as thyroglobulin. Facts observed by others can not be explained entirely on this basis.

Lunde, Closs and Pedersen[1] have found iodine to be present in normal blood in two forms. Part of it is precipitated with the proteins when these are thrown down by alcohol. Part is alcohol-soluble. The fraction that is precipitated by alcohol has been shown to be greatly above normal in the blood of patients suffering from Graves' disease, and to return to normal as the patients improve under treatment.[1] It is also greater than normal in the blood of experimental animals that have been injected with an extract of anterior pituitary.[2] The alcohol-soluble iodine does not vary much from the normal in either case. These findings, together with the fact that positive precipitin tests for thyroglobulin have been obtained in blood as it is leaving the thyroid,[3] indicate that the compound secreted is thyroglobulin. But, if this is so, it is difficult to understand how the thyroid hormone is able to affect the metabolism of all the cells of the body, for thyroglobulin is a highly indiffusible substance. Moreover, Dodds, Lawson and Robertson,[4] on examining the blood of a large number of patients suffering

[1] G. Lunde, K. Closs and O. C. Pedersen, *Biochem. Zeit.*, 206: 261, 1929.

[2] K. Closs, L. Loeb and E. M. MacKay, *Jour. Biol. Chem.*, 96: 585, 1932.

[3] A. J. Carlson, L. Hektoen and R. Schulhof, *Amer. Jour. Physiol.*, 71: 548, 1925.

[4] E. C. Dodds, W. Lawson and J. D. Robertson, *Lancet*, 2: 608, 1932.

from various types of thyroid disturbance, were unable to find any quantitative relation between the alcohol-insoluble iodine of the blood and the activity of the thyroid as measured by metabolic rate. This suggests the possibility that iodine may be secreted in more than one form.

To obtain further light on this subject, 20 cat and 8 rabbit thyroids were cultivated in the Lindbergh perfusion pump.[5] Then the media in which they were cultivated were fractionated and the various fractions analyzed for iodine.[6] The globulins were precipitated with ammonium sulfate, and the total protein fraction with alcohol. Control fluids were saved in each case and analyzed in the same manner as the perfusion fluids so as to correct for the iodine present in the medium before cultivation. Serum at 40 per cent. concentration in Tyrode's solution was used as the medium in most instances, though 20 and 80 per cent. serum were also tried. The perfusion time was varied from 4 hours to 6 days.

In every instance iodine that was set free by the gland was found in the medium in two forms. Part of it was precipitated with the globulins, and part was found in the globulin-free filtrate. Part was likewise found in the alcoholic precipitate and part in the alcoholic filtrate. The amount that was precipitated from any one medium was the same no matter which precipitant was used. And the sum of that recovered in the precipitate and that found in the filtrate was the same as that found in the unfractionated medium. Thus, gland No. 289 set free 25.2 gammas of iodine. 9.7 gammas of this was found in the precipitate, and 15.6 gammas in the filtrate. As is seen in this example, the iodine that is recovered in the filtrate is no small part of the total secreted. In some experiments it amounted to as much as 80 per cent. of the total, and as much as 40 per cent. of that originally present in the gland.

Presumably, the iodine that is found in the globulin fraction of the medium is contained in thyroglobulin or slightly modified thyroglobulin. That recovered in the filtrate is probably contained in hydrolytic products of the thyroglobulin molecule. It can not be derived from inorganic iodine stored within the gland, for the glands do not contain any such quantity of inorganic iodine. To make certain that this was the case, a number of uncultivated thyroids were fractionated and analyzed. The non-globulin iodine of the glands was found to vary with the total iodine content and constituted only 10 per cent. of the total. That found in the filtrate was often as much as 40 per cent. of the total.

[5] C. A. Lindbergh, *Jour. Exp. Med.*, 62: 409, 1935; A. Carrel, *Jour. Exp. Med.*, 65: 515, 1937.

[6] The method used was a modification of that described by V. Trevorrow and G. J. Faschena, *Jour. Biol. Chem.*, 110: 29, 1935, and 114: 351, 1936.

Because iodine appears in the medium in two forms it is not necessary to conclude that it is actually secreted in two forms. It is conceivable that thyroglobulin might be secreted and hydrolyzed later as cultivation is continued. And, as a matter of fact, a larger portion of the secreted iodine was found in the filtrate in experiments in which the glands were cultivated for 6 days than in those in which they were cultivated for only one day. Yet, when thyroglobulin extracted from cat thyroids was incubated with 40 per cent. serum for two weeks at the various pH values which exist during cultivation no hydrolysis occurred. Apparently, therefore, thyroglobulin is broken down only when the gland is present. To avoid changes that might occur after secretion had taken place experiments of very short duration were made. But whenever any measurable amount of iodine was secreted a part of it was always found in the filtrate. And, when thyreotropic hormone was added to the medium to stimulate secretion, more of the iodine secreted as the result of adding the hormone was recovered in the filtrate than was found in the precipitate. These facts seem to indicate that the iodine is actually secreted in two forms. The increased proportion found in the filtrates in long-continued experiments indicates that the gland continues to act on thyroglobulin that has been secreted as it is repeatedly returned to the gland in the circulating medium.

But if the thyroid secretes iodine in more than one form when it is cultivated outside the body, why has no evidence of such activity been obtained within the animal organism? There is, of course, the possibility that the gland functions differently *in vitro* than it does *in vivo*, and that glands of different animal species function differently. But the most plausible explanation seems to be that the smaller fragments of the thyroglobulin molecule, being more diffusible than thyroglobulin itself, are quickly absorbed by the various cells and tissues within the body and so do not remain in the blood stream.

To summarize: Iodine set free by the thyroid gland during its cultivation in the Lindbergh pump is recovered in the medium in two forms. Part of it is precipitated with the globulins of the medium, and part is found in the globulin-free and also in the protein-free filtrate. Evidence is given which indicates that the latter is contained in degradation products of the thyroglobulin molecule formed as the result of activity of the gland.[7]

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[7] Thanks are due Dr. Carrel and several of the members of his technical staff for cultivating the glands, and assistance in other parts of the work.