

incorporation is similar to that described by Herve and Chapeville and is clearly distinguishable from the alkali-labile incorporation described above. Thus, an incorporation system with substantial amounts of phenylalanine would show mainly NH₂-terminal incorporation of Phelac, instead of polyester formation. It is likely that this explains the difference between our results and the work reported by Herve and Chapeville.

We have attempted to measure the chain length of the polyester by a method closely related to that of Herve and Chapeville (8). In their experiments, ¹⁴C-labeled phenyllactate is attached to ³²P-labeled tRNA. In the absence of GTP, the ratio of ¹⁴C to ³²P is measured to assay the ribosomal bound Phelac-tRNA in the absence of polymerization. In the presence of GTP, polymerization ensues, and the ratio increases when more than one ¹⁴C residue is bound to the tRNA in the nascent polymer. If nonspecific binding of Phelac-tRNA occurs even in the presence of GTP, then this measurement gives a lower limit for the actual polymer length. The results of Herve and Chapeville showed no increase in the ratio in the presence of GTP. This was interpreted to indicate the presence of only one Phelac residue per polypeptide. We have repeated this measurement (9) and find that GTP induces an increase in [¹⁴C]Phelac bound of 14.7-fold relative to that bound in the presence of the analog GMP-PCP. This shows that polymerization has occurred, but in view of the possibility of nonspecific binding of tRNA to the ribosome, this number should not be interpreted quantitatively.

In most of these experiments the incorporation of ¹⁴C radioactivity into insoluble precipitate was less than 10 percent of the input radioactivity. It is likely that the incorporation is limited because of racemization and rearrangement of the amino acid which occurs during nitrous acid deamination of phenylalanyl-tRNA. In analogous experiments with nitrous acid deamination of puromycin (1), it was found that the majority of the deaminated material had undergone rearrangement, leaving only a small proportion with the original active configuration. The mechanism of these rearrangements has been studied in detail in simpler molecules (10).

In our work with puromycin analogs, we showed that the ribosomal peptidyl

transferase was able to form ester bonds as well as amide bonds (1). In an extension of this work we used the natural messenger RNA found in bacteriophage R17 and showed that the incorporating system was able to insert α -hydroxy acids into unique positions in a coat protein fragment (3). Our results here demonstrate that the ribosome can form consecutive ester bonds and produce a polyester when it has as a substrate α -hydroxyacyl tRNA. Analysis of the product suggests some incorporation of phenylalanine into the polyesters so that the product might be more properly described as a mixed polyester-amide. However, it is likely that incorporation of phenylalanine in the system could be eliminated entirely by fractionating the incorporating system and eliminating the peptidases which generate phenylalanine during the course of the incubation.

Demonstration of the fact that the ribosomal peptidyl transferase can utilize an α -hydroxyl residue as well as an α -amino residue in the synthetic reaction may have some bearing on the possibility that peptidyl transferase catalyzes the hydrolytic reaction in chain termination (1, 3). In addition it may be related to the fact that peptidases

which cleave peptide bonds also have the ability to cleave ester bonds. The specificity for both the synthetic and the hydrolytic reactions is similar. This similarity may be of value in understanding the mode of action of peptidyl transferase, an enzyme that occupies a central role in molecular biology.

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12. Supported by grants from NIH, NSF, and NASA.

16 March 1971; revised 21 May 1971

Sex Difference in the Number of Sympathetic Neurons in the Spinal Cord of the Cat

Abstract. *Counts of preganglionic sympathetic neurons in the spinal cords of four male and four female cats were obtained. The number of these neurons in males was significantly greater than that in females. These data suggest that the numbers of neurons in the mammalian central nervous system are different in the two sexes.*

It is well established that the mean weight of the brain of men is greater than that of women and that brain weight in the two sexes is poorly correlated with body weight and length (1). A similar sex difference has been demonstrated for the weight of the human cerebellum (2) and for the volume of the spinal cord in cats (3). At least four factors must be considered in attempts to account for these differences in weight and volume: (i) a difference in the number of neurons (4), (ii) a difference in their size, (iii) a difference in the number of glial cells (5), and (iv) a difference in the development of dendrites, which is usually expressed as the ratio of neuropil to perikaryon (6).

We now report data demonstrating that the number of sympathetic preganglionic neurons in the spinal cord of the cat is greater in the male than in the female. In the course of an investigation of descending autonomic connections between the medulla and the spinal cord it became necessary to study the numerical distribution of neurons in the thoracolumbar intermediolateral nucleus (ILN) of the spinal cord, and the results presented here are part of a larger study to be published elsewhere (7).

Eight adult cats (2.2 to 3.0 kg) were anesthetized and perfused with 0.9 percent saline and then with 10 percent formol in saline. The spinal cords were removed and fixed in 10 percent for-

Table 1. Estimated number of sympathetic neurons in the thoracolumbar intermediolateral nucleus of cats of both sexes. The mean (and standard error of the mean) weight and number of neurons for female cats were 2.75 ± 0.19 kg and $35,543 \pm 1,411$, respectively. For male cats the mean weight was 2.63 ± 0.13 kg; the mean number of neurons was $45,765 \pm 2,556$.

Cat number	Weight (kg)	Neurons (No.)
<i>Female cats</i>		
9	3.0	32,790
12	2.2	37,570
13	3.0	38,350
15	2.8	33,460
<i>Male cats</i>		
11	2.6	53,340
14	3.0	43,300
16	2.5	42,170
17	2.4	44,350

mol in saline for at least 2 weeks. Each spinal cord, trimmed to include segments C₆ to L₅, was divided into four parts which were embedded in paraffin. Serial transverse sections 15- μ m thick were cut, and every tenth section was mounted and stained with thionine (8). A total of 9400 sections was studied at a magnification of $\times 320$. We identified ILN neurons by well-defined criteria (9), and estimated their total number by multiplying by 10 the number of nucleoli counted in the ILN of both sides. We assessed the accuracy of this method of counting by comparing the estimated count with the actual count of nucleoli in a series of 200 consecutive serial sections in one specimen; the numbers of counted and estimated nucleoli were 2022 and 1980, respectively.

The ILN extends from the junction of segments C₈ and T₁ to L₄ and is located within the lateral horn of the gray matter. Total neuron counts, body weights, and sexes of the eight animals studied are shown in Table 1. The mean counts for males and females are significantly different ($t = 3.5$, $P < .02$), although no significant difference is apparent between the mean weights of the two groups ($t = 0.54$, $P > .9$).

The data presented demonstrate a sex difference in the number of preganglionic sympathetic neurons in the cat. We are not aware of a sex difference in the number of central neurons having been reported before. However, this finding does not necessarily imply that the total number of neurons in the central nervous system is greater in the male. This sex difference may be limited

to the sympathetic nervous system as it has been demonstrated that the number and size of neurons in sympathetic ganglia may be increased by the administration to newborn mice of a nerve growth factor isolated from the submaxillary gland of male mice; the amount of this factor present in the gland is decreased by ablation of the testes and increased by administration of testosterone to female rats (10). In addition, as it has been demonstrated that the development of chick dorsal root ganglia is influenced by the size of the tissue innervated (11), a possibility to be considered is that the sex difference in the number of ILN neurons may be secondary to a sex difference in the size of the viscera supplied by the sympathetic nervous system. Because no information is available on the size in the two sexes of organs supplied by the sympathetic nervous system, this possibility cannot be excluded. Finally, because no information was available on the age of these animals, it is possible that a difference in the body weights of cats of the two sexes was not demonstrated because some of the animals in our sample were not fully developed. This possibility is given substance by the demonstrated sex difference in weights of adult cats (12). The interpretation of our results would then be that the greater number of ILN neurons in the male is related to the statistical expectation of a greater body mass in the fully developed animal. However, whether or not this difference in the number of ILN neurons is related to body or visceral mass as well as to sex, a confirmation of our findings in other areas of the central nervous system would raise interesting questions

regarding the influence of genetic and environmental factors on the developmental dynamics of neurogenesis in the two sexes.

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9. The neurons of the ILN are either bipolar or multipolar and are usually pyriform in shape with the long axis two to three times the length of the short axis. Their nuclei are located in a central position and contain darkly stained nucleoli. The scanty cytoplasm contains Nissl substance arranged in packets of irregular size and shape. The distinction between neurons and glial cells is always unequivocal because neurons contain nucleoli clearly surrounded by nucleoplasm. Neurons of the ILN are easily differentiated from those in neighboring structures at the lateral and dorsal borders because of intervening white matter. The ventral border is usually distinct because ILN and ventral horn neurons differ in size and are clearly separated. The medial border, however, is occasionally difficult to establish because of the presence of scattered neurons between the ILN and the intermediomedial nucleus; in these cases only those neurons which clearly appear as part of a group of other ILN cells are included.
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13. Supported by the Medical Research Council of Canada (grant MT 2096). We thank Mr. M. R. Thomas for bringing to our attention the possibility of a sex difference, and Mrs. C. J. Kuo for her technical help.

26 April 1971; revised 4 June 1971

Glycogen Plastids in Müllerian Body Cells of *Cecropia peltata*—A Higher Green Plant

Abstract. *A glycogen-containing plastid has been found in the cells of Müllerian bodies on Cecropia peltata (Moraceae) trees. Plastids in cells of the leaf blade, petiole, and stem are of the usual chloroplast type and contain starch grains only. Müllerian bodies exist for the purpose of providing a food supply for a protective, mutualistically inhabiting ant population. A series of chemical tests and observations indicates that this glycogen is identical to animal glycogen.*

Traditionally there has been a sharp distinction in the terminology used to describe the method of glucose accumulation and storage between plants and animals. Higher plants have been found

to store their excess glucose in the form of starch, whereas animals produce a polysaccharide termed glycogen. Starch usually takes the form of a grain, a few microns in diameter, and is located in