APPEARANCE OF GLYCOGEN AND NISSL'S BODIES IN THE NERVE CELLS OF THE CHICK EMBRYO

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By

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Introduction

It has been generally known that the so-called chromatolysis, wholly or in part, occurs in the nerve cells under various pathological conditions. Previously it was reported by Toryu (1938) that the disintegration of Nissl's bodies in the nerve cells of guinea-pigs after a section of their axones is accompanied by glycogenolysis, but when the recovery of the Nissl's bodies takes place the glycogen granules of the same form and arrangement as Nissl's bodies reappear, showing that the glycogen in the nerve cells is contained in Nissl's bodies. Later the same author (1949) also found the same relation in the nerve cells of horses affected with paralysis caused by *Microfilalia* and of hens affected with B, avitaminosis.

In the present investigation I have dealt with the appearence of the glycogen and Nissl's bodies in the nerve cells of the chick embryo during their development with the hope of obtianing further data concerning the evidence of the presence of glycogen in the Nissl's bodies.

Materials and Methods

All the materials were taken from the embryos of White Leghorn. The newly laid eggs were kept in an incubater of ordinary type approximately at 39°C under moderate ventilation and moisture. The development of the embryo resulted without injury throughout the entire period of incubation.

The entire bodies of the embryos at various stages of development were fixed in alcohol-formalin saturated with sodium acetate (Toryu). The trunk regions, in which the spinal cord and the spinal ganglia were contained, were taken from the fixed embryos and used for the histochemical analysis of the relation between the glycogen granules and Nissl's bodies.

All the materials were embedded in celloidin and sectioned 15μ thick.

The following stains were used: Methylene blue or toluidin blue for Nissl's bodies, Best's carmine fluid for glycogen.

Results

1. *Result Obtained for the Morphological Relation between Nissl's Bodies and the Glycogen Granules in the Nerve Cells of the Adult Fowl.*

I have preliminarily studied the morphology of the glycogen granules in the nerve cells of the adult fowl, with reference to the relation of it to Nissl's bodies. The observation is as follows:

In the large moter cells in the anterior horn of the spinal cord the glycogen granules are of angular form of conspicuous size, which often exceed in dimension the large nucleolus of the nerve cell. They are to be found not only in the cell body, but also for a distance in the dendrites, where they have a more elongated, spindle shape. The neurite and its cone of origin are free from them. Sensory cells in the posterior horn of the spinal cord contain a few large spindle shaped granules.

In the cells of the spinal ganglion, the glycogen granules are in an irregular roundish form of large size and closely packed in the body of the cell. It was also found, by the method stated in 1937 by the present author, that the glycogen granules just stated are morphologically the same as Nissl's bodies, showing that the glycogen is contained in the nerve cells at least as one of the elements of Nissl's bodies. This confirms the result obtained for the glycogen in the nerve cells of the horse (1937) and of the guinea-pig (1938).

2. *Result Obtained for the Appearance of Glycogen.*

The appearance of glycogen in the nerve cells of the spinal ganglion and the spinal cord of the chick embryo at various stages of development was morphologically examined, and the results are as follows:

A. *Appearance of Glycogen in the Spinal Ganglion.*

In the embryo of about from 2 to 3 days the neural crests, the rudiments of the spinal ganglia appeared between the neural tube and the surface ectoderm. The cells of the neural crests contained no glycogen.

In the embryo of about from 4 to 5 days the neural crests were broken into a metameric series of cell-groups containing neuroblasts and situated laterally between the ectoderm and the mesodermal somites. The cells in the ganglion thus formed were small and somewhat elongated or irregular in shape and contained a trace of glycogen, though it was not definitely demonstrated as yet.

During the 6th day the neuroblasts showed marked modifications; they

increased in size and sent out their axis cylinder prccesses, aquiring the typical form of the ganglion cells, though their size was much smaller than the normal cells. In these cells glycogen was found, but not closely packed as yet in the cell bodies (Fig. 1). In some cells the glycogen granules appeared only in the periphery, where the granules were so small and so closely packed as to make the cells dusty.

Succeeding stages showed the accumulation of glycogen; namely, with the procedure of incubation the nerve cells became large and the glycogen granules increased in both their size and amount and until after about from 13 to 16 days of incubation their form and arragement were found to be normal (Fig. 2).

B. *Appearance of Glycogen in the Spinal Cord.*

The appearance of glycogen in the spinal cord was closely related with that in the spinal ganglion. During the sixth day of incubation the wall of the cord greatly thickened and the glia cells and the neuroblasts or nerve cells were differentiated from the germinal cells, especially in the ventro-lateral wall where relatively large motor cells having axis cylinder processes were formed. In these nerve cells glycogen granules were found poorly stained with Best's carmine. They were spindle or roundish in form, of small size and very few in number. As the incubation proceeded their size increased and finally in about from 13 to 16 days of incubation, when well marked motor cells and sensory cells were found, the glycogen granules of the same form and arrangement as those found in the adult stage appeared (Fig. 6).

It was also found that the appearance of glycogen in the nerve cells far precedes those in the heart muscles and the liver cells, in the heart muscles the first appearance of it being about the eighth day and in the liver cells about the thirteenth day.

3. *Result Obtained for the Relation of Glycogen to Nissl's Bodies.*

Preceding the investigation concerning the relation of glycogen to Nissl's bodies, the appearance of the bodies were morphologically examined. The observations are as follows:

In about the 6th day of incubation Nissl's bodies first appeared as fine granules in the nerve cells of the spinal cord and spinal ganglion. They increased in size and in amount with the process of incubation up to about 13 to 16 days, when Nissl's bodies were almost completed, reaching the normal state of the adult fowl. This shows that the appearance of Nissl's bodies always agrees with that of the glycogen granules mentioned in the previous section.

To determine whether or not the appearance of glycogen in the nerve

cells of the chick embryo is also related to the appearance of Nissl's bodies, the sections from the embryos at various stages of development were used for both the Nissl staining and glycogen staining by the method stated in 1937. The microscopic examination was made on the sections thus treated and Nissl's bodies were morphologically compared with the glycogen granules. The observation is as follows:

Stained by Best's carmine fluid after decolorization of Nissl's bodies, red colored granules appeared and the form and the arrangement of them were found to be strictly the same as those of Nissl's bodies (Fig. 3-6). It was also found that the degree of the glycogen staining always shows a good agreement with that of Nissl's staining, indicating that both the glycogen and Nissl's bodies always appear in the same time and also in the same portion of the cells.

Kimura (1934) studied the glycogen in the central nervous system of the vertebrates fixed in Carnoy's solution and stated that in the adult it is found intracellularly in Nissl's bodies, but in the embryonal stage it occurs principally extracellularly. In the present investigation of the nerve cells of the chick embryos fixed in alcohol-formalin saturated with sodium acetate (Toryu), however, it was found intracellularly.

At any rate, from the results above obtained it is highly probable that the glycogen granules contained in the nerve cells are the same as Nissl's bodies when the morphological relation alone is considered or that the glycogen is contained in the nerve cells as one of the elements of Nissl's bodies when not only the morphological relation, but also the componental relation are considered. To support the view just stated I notice the following facts; first, the disappearance of Nissl's bodies in the nerve cells of the horse by the action of digestive enzyme or by a post-mortem autolysis shows the disappearance of glycogen, as has already been stated by the present author in 1937, and second, the so-called chromatolysis occurring in the nerve cells of guinea-pigs after a section of their axones is accompanied by glycogenolysis, in 1938, and third, in horses and fowls affected with paralysis of their nervous systems both the chromatolysis and glycogenolysis occurs in the same manner as above mentioned, in 1949.

Summary

The results obtained in this investigation may be summarized as follows:

1. The glycogen in the nerve cells of the chick embryo began to appear in about the 6th day of incubation and reached the normal state in about from 13 to 16 days.

2. The appearance of Nissl's bodies agreed with that of the glycogen.

3. The glycogen granules in the nerve cells were morphologically coincident with Nissl's bodies; they could be stained with Best's carmine fluid or methylene blue.

Literature Cited

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Explanation of Figures

Fig. 1. Glycogen in the spinal ganglion cells of the chick embryo of the 6th day of incubation. ×650.

Fig. 2. Glycogen in the spinal ganglion cells of the chick embryos after about 16 days of incubation. ×650. (Glycogen has almost completely appeared)

Fig. 3. Nissl's bodies in the spinal ganglion cells of the chick embryo of about 13 days of incubation. ×650.

Fig. 4. Glycogen in the same cells as the above. ×650. (Nissl's bodies were decolorized and then stained with Best's carmine fluid. The form and arrangement of the glycogen granules are the same as those of Nissl's bodies.)

Fig. 5. Nissl's bodies in the anterior horn cells (motor cells) of the spinal cord of the chick embryo of about 16 days. ×650. (Nissl's bodies have almost completely appeared.)

Fig. 9. Glycogen in the same cells as the above. ×650. (Glycogen has also completely appeared.)

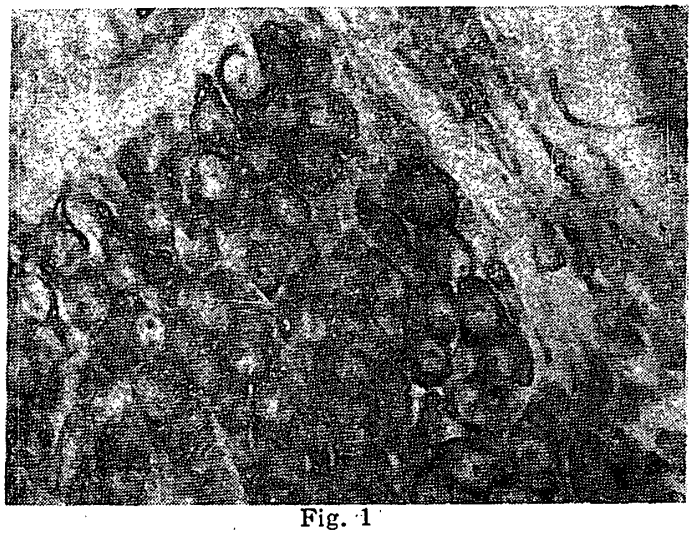


Fig. 1

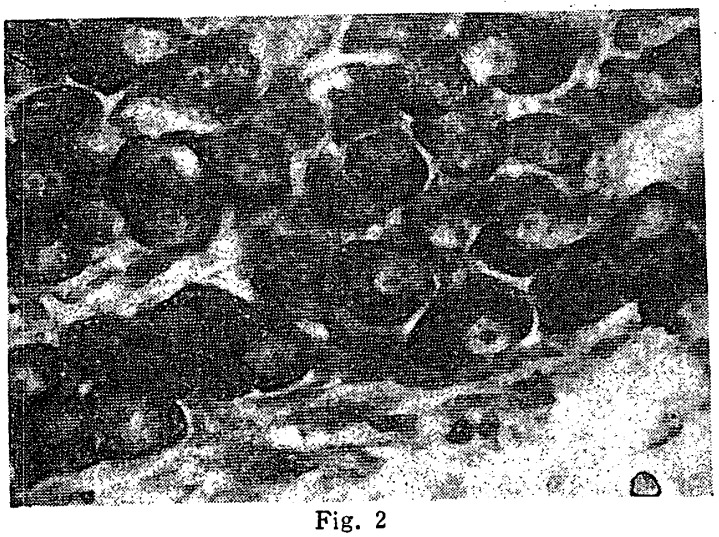


Fig. 2

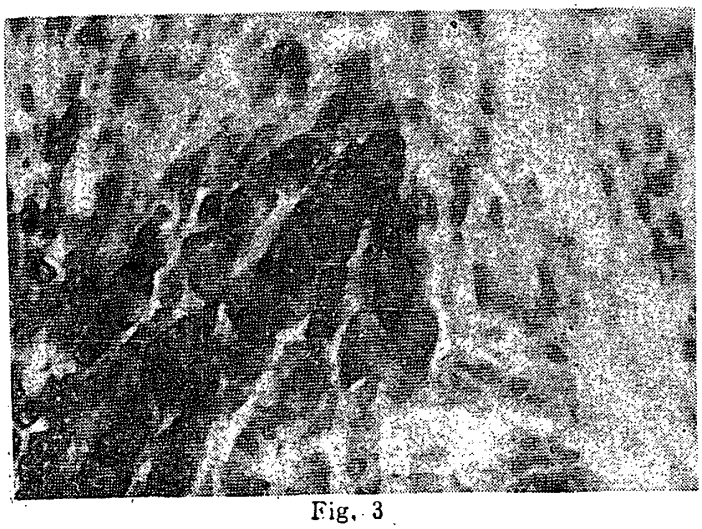
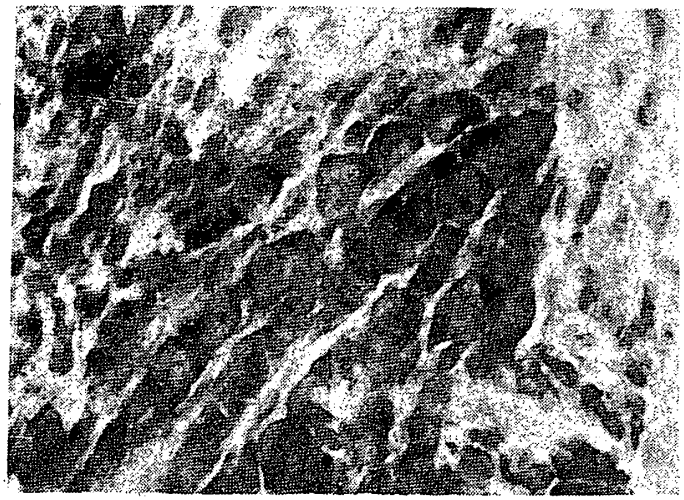


Fig. 3



Flg. 4

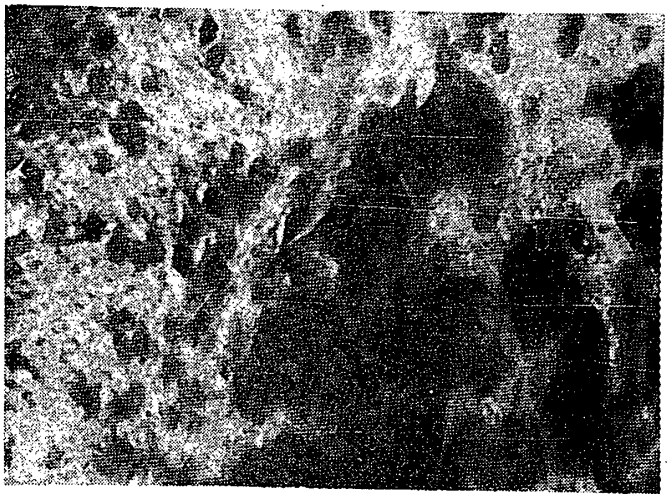


Fig. 5

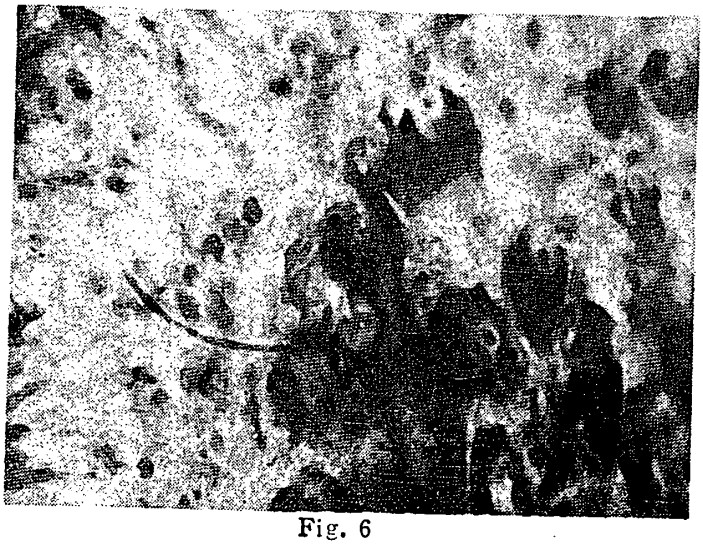


Fig. 6