CHANGES IN SIALIC ACID CONTENT IN THE INTESTINAL TISSUES OF ANIMALS VACCINATED WITH LIVING POLIO-MYELITIS VACCINE

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A single oral administration of living poliomyelitis vaccine of immunologic type II (Sabin strain) to albino mice weighing 12-14 g is followed by an increase in the free sialic acid content from the 5th to the 20th days and in the bound sialic acid content from the 20th to the 45th day from the beginning of immunization in the small intestine of the vaccinated animals.

KEY WORDS: poliomyelitis; immunization with living poliomyelitis vaccine; sialic acids.

Several workers have expressed the view that an increase in the serum sialoprotein level is one of the mechanisms of nonspecific resistance of the living organism [2-4, 6]. Investigations by the writers have shown that oral immunization with living poliomyelitis vaccine causes a prolonged increase in the sialoprotein level not only in the blood serum, but also in the lungs of the vaccinated animals [1]. Antigen of attenuated poliomyelitis virus has been found by the fluorescent antibody method in the lungs of these animals. The suggestion has been made that changes in the sialoprotein level in the lungs are the result of direct action of the virus on the tissue. The point of primary application and reproduction of poliomyelitis virus is known to be the intestine.

In this investigation the content of free and bound sialic acids was studied in the tissues of the small intestine of animals vaccinated orally with living poliomyelitis vaccine.

EXPERIMENTAL METHOD

A single dose of $0.5\,\mathrm{ml}$ (a massive dose) of living poliomyelitis vaccine of immunologic type II (Sabin strain), with a titer of 10^{-7} CPD₅₀/ml measured on monkey kidney cells, was given by mouth to albino mice weighing $12\text{--}14\,\mathrm{g}$. The animals of the control group were given a placebo, which contained all the ingredients of the vaccine except the virus. The content of specific antibodies in the blood of animals immunized with living poliomyelitis vaccine was determined in the neutralization test on a culture of primary trypsinized monkey kidney cells.

The content of free and bound sialic acids in digests of intestinal tissue, preliminarily washed with 0.1 M phosphate buffer to remove blood, was determined by Aminoff's [5] method. At each period 10 albino mice were killed. The experiments were repeated six times.

To determine free sialic acids in the tissues a weighed sample (1 g) of the organs was homogenized with 3 ml of 2% TCA solution. The sample was centrifuged for 10 min at 3000 rpm and 0.5 ml of supernatant was used for the experiments. To determine sialoproteins, a sample of organs weighing 1 g was homogenized with 3 ml 5% TCA and hydrolyzed for 20 min

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TABLE 1. Content of Free and Bound Sialic Acids (in $\mu g/g$ wet weight of tissue) in Tissues of Small Intestine of Albino Mice Immunized Orally with Living Poliomyelitis Vaccine (M \pm m)

Substance determined	Time of investigation (in days)						
	õ	7	14	20	30	45	60
Free sialic acids: experiment control P Bound sialic	90,0±4,0 75,0±4,2 <0,05	94,0±9,4 69,0±2,2 <0,05	167,0±10,1 102,0±7,4 <0,01	157,0±6,3 92,0±12,3 0,01	66,0±4,2 68,0±1,6	69,0±6,1 61,0±2,9	81,0±9,2 71,0±14,2
acids: experiment control P	974,0±32,5 1011,0±27,5 —	1084,0±18,4 967,0±65,9	843,0±54,7 756,0±20,6 —		1230,0±95,4 812,0±124,2 <0,05		773,0±94,1 747,0±95,2 —

on a boiling water bath to rupture the α -ketoside bond uniting the sialic acids with the protein moiety of the sialoprotein molecule. After hydrolysis, 0.5 ml of the filtered digest was used for the experiment.

EXPERIMENTAL RESULTS

The sialic acid level in the intestinal tissues was determined in the course of 2 months after immunization with living poliomyelitis virus.

Investigation of the dynamics of the sialic acid content of the small intestine of the vaccinated animals showed that in the early stages of the immunologic response the free sialic acid content was considerably increased. From the 5th to the 20th day there was a significant increase in the content of these compounds P < 0.05). Later the content of free sialic acids fell to normal, and throughout the subsequent period of observation no significant abnormality was found.

Meanwhile, starting from the 20th day after the beginning of immunization, the sialoprotein content of the tissues of the small intestine of the vaccinated animals was significantly increased. The content of these compounds remained high until the 45th day (P < 0.05). By the 60th day the sialoprotein level was again close to the control value (Table 1).

The increase in the content of free sialic acids in the intestine of animals vaccinated with poliomyelitis vaccine always preceded the time of the highest blood level of specific antibodies. For instance, the serum antibody titer on the 15th, 20th, and 30th days after oral administration of the vaccine was 1:16, and on the 45th and 60th days it rose to 1:64.

The results suggest that during the formation of postvaccinal immunity to poliomyelitis the attenuated poliomyelitis virus, which propagates in the small intestine, has a marked effect on the sialic acid level in the cell and in its structures. The increase in the content of free sialic acids in the intestine is possibly due to activation of lysosomal enzymes containing cellular neuraminidase. The increase in the sialoprotein level in the intestine can evidently be regarded as one of the local factors of the nonspecific resistance of the organism.

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