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**Key words:** major salivary glands, parenchymal cells, stromal cells, intrauterine antigenic action, glycosaminoglycans, the rats.

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## DISTRIBUTION FEATURES OF THE RATS' MAJOR SALIVARY GLANDS CELLS GLYCOPROTEINS DURING EARLY POSTNATAL PERIOD AFTER ANTENATAL ANTIGEN ACTION

*This study was done within the research «Lectin histochemical characteristics of morphogenesis of the organs and tissues in early postnatal period in norm and experiment» (state registration number 0109U003986).*

**ABSTRACT. Background.** Nowadays, in the diseases' structure, according to literature data, one of the leading place take pathological condition connecting with the salivary glands' inflammatory and dystrophic disorders. The problem of etiology and pathogenic not enough studied and demanded intent attention of researchers. **Objective.** The purpose was to determine the features of glycoproteins' distribution in the structures of rats' major salivary glands in early postnatal period after intrauterine antigen action. **Methods.** The object of the research was 224 salivary glands of white laboratory rats. Due to impossible quality materials' taking in the early periods of postnatal life parotid and sublingual salivary glands, the investigation done at the gl. submaxillaris. The histochemical exposure and differentiation of carbohydrate compounds conducted by means of PAS-staining technic. For fermentative control used diastase. The results of histochemical exposure of glycoproteins stain were done by semi-quantitative. **Results.** In newborn animals receiving antigen in the antenatal period, in the cells' cytoplasm indicate the accumulation' increase of PAS-positive compounds retained until the 14th and offset at the 45th day of postnatal life. The detected changes in the major salivary glands cells' are the basis for the development of inflammatory and dystrophic processes and can lead to the functional violations formation' hereinafter. **Conclusion.** Our findings indicate that at the background of intrauterine antigen action, the glycoproteins' accumulation intensity in parenchymal and stromal cells' cytoplasm of the major salivary glands is decrease, but glycogen content is increase compared with intact animals group. Furthermore, we detected cells' secretory activity reduction from 1<sup>st</sup> to 14<sup>th</sup> day of postnatal life with increase glycogen' accumulation in the cells' cytoplasm. That revealed changes offset at the 45<sup>th</sup> day after birth in all animals group.

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**Сирцов В.К., Маслова І.М. Особливості розподілу глікопротеїнів в клітинах великих слинних залоз щурів в ранньому постнатальному періоді після антенатальної антигенної дії.**

**Реферат.** Мета роботи – встановити особливості розподілу глікопротеїнів в клітинах великих слинних залоз в ранньому постнатальному періоді після внутрішньоутробної антигенної дії. У новонароджених тварин, що отримали антиген в антенатальному періоді, в цитоплазмі клітин виявлено зниження накопичення ШЙК-позитивних сполук, яке зберігається до 14-ї та нівелюється на 45-ту добу. Виявлені зміни в клітинах великих слинних залоз є підґрунтям для розвитку запальних та дистрофічних процесів в слинних залозах, що, в подальшому, може призвести до виникнення різних функціональних порушень.

**Ключові слова:** великі слинні залози, клітини строми, клітини паренхіми, внутрішньоутробна антигенна дія, глікопротеїни, щури.

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### Introduction

Nowadays, in the diseases' structure, according to literature data, one of the leading place take pathological condition connecting with the salivary

glands' inflammatory and dystrophic disorders. The problem of etiology and pathogenic not enough studied and demanded intent attention of researchers [1]. One of determinatives that result in violation of

major salivary glands morphogenesis of and as a result the development of its pathology is the condition of pregnant health, more than half of them has chronic diseases and system functional disorders which is accompanied by the immune pathological condition, namely by antigen influence on a fetus [2]. Nowadays it is determined injection of the antigen in the antenatal period is the basis for the development of inflammatory processes in a postnatal period [3]. Interconnection of glycoproteins (GP) distribution in the major salivary glands structures in a postnatal period injected intrauterine an antigen after birth is not enough studied. Therefore, it is necessary to investigate in detail content and distribution of carbohydrate compounds in the structures of rats' major salivary glands after an intrauterine antigen action.

Furthermore, the sugar – containing proteins play one of the main role in the salivary glands' metabolic process. However, it remains to elucidate whether these energy sources used for secretion, excretion or both. In addition, the differences in the function of excretion and the role of the secretory cells are currently unknown in salivary glands at the background of intrauterine antigen action.

#### **Purpose**

To determine the features of glycoproteins' distribution in the structures of rats' major salivary glands in early postnatal period after intrauterine antigen action.

#### **Methods**

The object of the research was 224 salivary glands of white laboratory rats. Due to impossible quality materials' taking in the early periods of postnatal life parotid and sublingual salivary glands, the investigation done at the gl. submaxillaris (in accordance with Nomina Anatomica Veterinaria, 2005).

The rats were divided into 3 groups. The 1<sup>st</sup> group – intact rats. The 2<sup>d</sup> group – rats, which were introduced 0,05 ml solution of antigen in the amniotic fluid on the 18<sup>th</sup> day of pregnancy, the 3<sup>d</sup> group – control, the animals were introduced intrauterine 0,05 ml of physiological solution on the 18<sup>th</sup> day of pregnancy. The feeding of animals was carried out twice a day at the same time.

For the study of peculiarities of GP distribution of the structures of major salivary glands of antigen's action on the fetus, was chosen the model of transuterine, transmembrane introduction of antigen in amniotic waters. As antigen was chosen rare (killed) split-vaccine Vaxigrip 2009. Keeping the animals and experiments were carried out accordingly to regulations of European convention about the defense of spine animals which are used due to the experimental and other scientific aims (Strasbourg, 1986), general ethic principles of the experiments on the animals taken by the first national congress of Bioethics (Kyiv, 2001). The animals' killing and taking of the material done from 13-00 till 14-00 on the 1<sup>st</sup>, 5<sup>th</sup>, 7<sup>th</sup>, 11<sup>th</sup>, 14<sup>th</sup>, 30<sup>th</sup>, 45<sup>th</sup> day of postnatal

life. On every term in all groups of the animals were examined 5 - 6 animals from 2-3 afterbirth. For the investigation, the major salivary glands used during some minutes after killing. The samples fixed in 10% solution of formalin, dehydrated, filled in paraffin mixture and produced serial paraffin sections. The histochemical exposure and differentiation of carbohydrate compounds conducted by means of PAS-staining technic. For fermentative control used diastase. The results of histochemical exposure of glycoproteins stain were done by semi-quantitative and determine as: +++ is the claret-red stain, ++ - pink-red, + is pinky, - - is absence of stain. Intermediate hues evaluated accordingly: ++/+++; +/++. Distribution of PAS - positive compounds studied in the major salivary glands' parenchymal and stromal cells' cytoplasm.

#### **Results**

At the first day of postnatal life for intact animals the glycoproteins accumulation in the parenchymal cells cytoplasm' more expressed than in experimental group - accordingly. Staining of all above-stated structures for the control group of animals does not differ from the data got from the animals of intact group, that is why in the future control group will not be cited (table 1).

After diastase fermentative control in the experimental group the stain intensity is -/+, in intact group is -. That showed the presents of glycogen' greater quantity in parenchymal cells' cytoplasm. We assume that the minor glycogens' quantity increase in the cells' cytoplasm at the 30<sup>th</sup> day in antigen-premium group can be connected with the cells' secretory process decompensation at the background. In experimental group, in difference intact animals the total quantity is decrease (table 1). In experimental animals group the glycogen' quantity remains increase compared with intact rats. That tendency saved to 7<sup>th</sup> day of postnatal life in all animals group. The 11<sup>th</sup> and 14<sup>th</sup> day characterized gradual content rise of glycoproteins' total quantity in intact group from +/++ to ++ and from + to ++ in experimental group relatively previous observation term. After diastase fermentative control the stain intensity in all animals groups is increased and shows the increasing intensity of synthetic processes, but the glycogen' quantity remains insignificantly increase.

At the 30<sup>th</sup> day after birth the glycoproteins' total quantity is ++/+++ in all animals group compared with previous observation term. The diastase digestion result to stain' intensity decrease in the experimental group as a consequence of glycogen' increase quantity. These findings indicate the synthetic and energy – intensive processes' deceleration. It is can be connected with attrition +; ++/+++ at the background of antigen action and animals' transition to the naturally type of feeding. At the 45<sup>th</sup> of postnatal life the stain intensity of parenchymal cells' cytoplasm is ++/+++ in all animals group.

Table 1

Glycoproteins' distribution in the parenchymal and stromal cells' cytoplasm of rats' major salivary glands

Day of post-natal life	Groups of animals	Parenchymal Cells		Stromal Cells	
		PAS	PAS+A	PAS	PAS+A
1	Intact	+/+++	+	+	-/+
	Experimental	+	-/+	-/+	-/+
	Control	+/+++	+	+	-/+
5	Intact	+/+++	+	+	-/+
	Experimental	+	-/+	-/+	-/+
	Control	+/+++	+	+	-/+
7	Intact	+/+++	+	+	-/+
	Experimental	+	-/+	-/+	-/+
	Control	+/+++	+	+	-/+
11	Intact	++	+/+++	+/+++	+
	Experimental	++	+/+	+/+++	+
	Control	++	+/+++	+/+++	+
14	Intact	++	+/+++	++	+/+++
	Experimental	++	+	++	+/+++
	Control	++	+/+++	++	+/+++
30	Intact	++/+++	+/+++	++/+++	+/+++
	Experimental	++/+++	+	++/+++	+/+++
	Control	++/+++	+/+++	++/+++	+/+++
45	Intact	++/+++	+/+++	++	+/+++
	Experimental	++/+++	+/+++	++	+/+++
	Control	++/+++	+/+++	++	+/+++

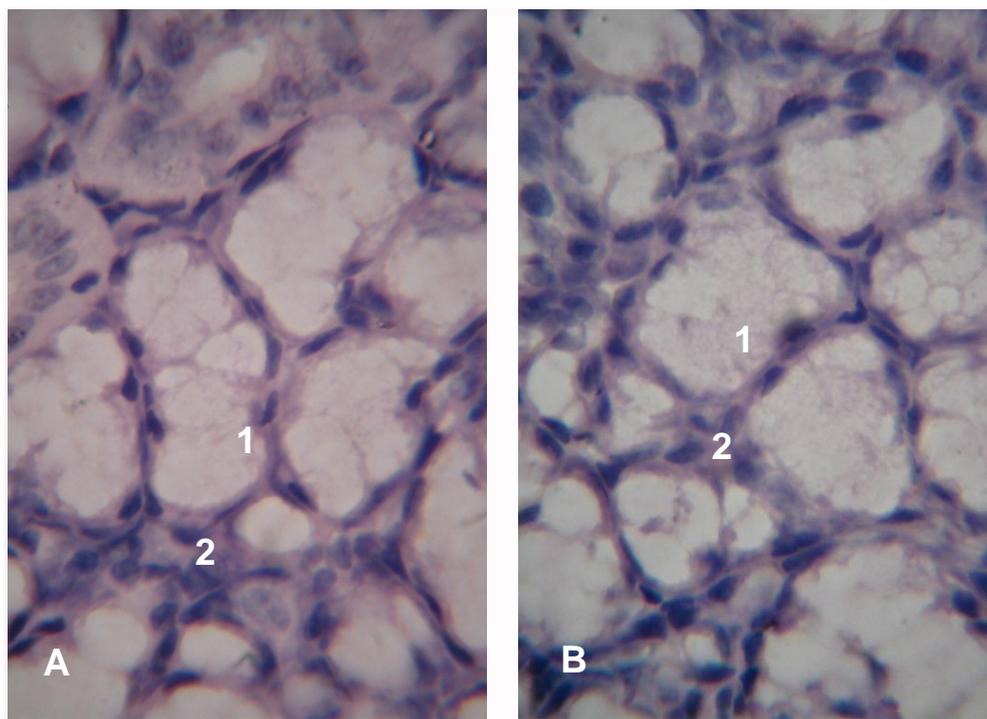


Fig. 1. Representative sections from rats' submandibular glands of experimental group, the 1 st day after birth (a – PAS reaction; b – PAS reaction with Diastase; Nuclei Coloration – Ehrlich's Hematoxylin; Original Magnification  $\times 1000$ ) 1 – parenchymal cells; 2 – stromal cells.

Besides the acinar structures, the major salivary glands contain the sufficient quantity of connective tissue. In the stromal cells' cytoplasm also detected glycoproteins' synthesis and accumulation imbal-

ance during the two weeks after birth. We identified the glycoproteins' total quantity decrease in experimental group on the background of glycogen' increase content relatively intact group. This imbal-

ance observed to the 14<sup>th</sup> day of postnatal life.

In the major salivary glands' stromal part the cells' cytoplasm contains the increase content of glycogen from 1<sup>st</sup> to 14<sup>th</sup> day of postnatal life and shows the reduce of cells' synthetic activity in experimental group. The detected changes of glycoproteins' and glycogen' synthesis and accumulation saved during two weeks after birth and offset at the 45<sup>th</sup> day of postnatal life in all animals group.

Thus, after fermentative control by using diastase solution at the 1<sup>st</sup>, 5<sup>th</sup>, 7<sup>th</sup> days after birth was detected the less intensive stain in experimental animals group compared intact group. That indicates the quantity changes of glycogen in major salivary glands cells' cytoplasm and shows the changes of cells' synthetic activity and energy – intensive processes.

### Discussion

In newborn rats after antigen' injection in amniotic fluid the intensity of the glycoproteins' accumulation in parenchymal cells' cytoplasm of major salivary glands is reduced in comparison with the intact animals group. This trend is growing in these cells to the 7<sup>th</sup> day. At the 11<sup>th</sup> and 14<sup>th</sup> day, the total number of glycoproteins aligned. The level of metabolism energy in these cells is higher than in intact and control animals and, due to the changes in proliferative activity. These changes are characteristic from the newborn period of the 14<sup>th</sup> day of life and practically disappeared on the 30<sup>th</sup> day.

In the study of stromal cells' cytoplasm observed intensity of PAS -positive compounds accumulation from the 1<sup>th</sup> to the 7<sup>th</sup> day. The glycoprotein' intensity accumulation increased in all groups of observations, but it is higher on the 7<sup>th</sup> day in antigen awarded animals group. It is marked the increase of PAS-positive compounds accumulation in early stages. From the 14<sup>th</sup> day the stromal structures cells' staining intensity reaches a maximum and re-

mains constant on the 45<sup>th</sup> day in intact and experimental groups.

The obtained results partially according to the changes of the PAS-positive compounds' accumulation, coincided with the data received early by several authors in the throat' mucous membrane, gums, pancreas and indicate to the synthesis imbalance of carbohydrate substances after antenatal antigen action [4; 5]. Insignificant difference among the data due to term' mismatching, absence of histogenetic affinity and morpho - functional specificity of major salivary glands. In addition, glycoproteins may be physiologically involved in an important part of the transporter system, not only in the acinar serous cells and the striated duct cells, but also in the excretory duct cells in the salivary glands [6]. Glycoproteins' reduction in the cytoplasm of stromal and acinar cells may be a reflection of nonspecific immunological protection it can lead to disruption of immunological barrier and be a basis for the development of inflammatory and dystrophic disorders of major salivary glands.

### Conclusion

Our findings indicate that at the background of intrauterine antigen action, the glycoproteins' accumulation intensity in parenchymal and stromal cells' cytoplasm of the major salivary glands is decrease, but glycogen content is increase compared with intact animals group. Furthermore, we detected cells' secretory activity reduction from 1<sup>st</sup> to 14<sup>th</sup> day of postnatal life with increase glycogen' accumulation in the cells' cytoplasm. That revealed changes offset at the 45<sup>th</sup> day after birth in all animals group.

### Further researches prospects

Given the imbalance of glycoproteins' and glycogen' synthesis and accumulation in the cells' cytoplasm appears the necessity to study the glycosaminoglycans' distribution peculiarities in the rats' major salivary glands.

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**Сырцов В.К., Маслова И.Н. Особенности распределения гликопротеинов в клетках больших слюнных желез крыс в раннем постнатальном периоде после антенатального антигенного действия.**

**Реферат.** Цель работы – установить особенности распределения гликопротеинов в клетках больших слюнных желез крыс в раннем постнатальном периоде после внутриутробного антигенного действия. У новорожденных животных, получивших антиген в антенатальном периоде, в цитоплазме клеток больших слюнных желез выявлено снижение накопления ШИК-позитивных соединений, которое сохраняется до 14-х и нивелируется на 45-е сутки. Выявленные изменения в клетках больших слюнных желез являются основой для развития воспалительных и дистрофических процессов в слюнных железах, что, в последующем, может привести к возникновению различных функциональных нарушений.

**Ключевые слова:** большие слюнные железы, клетки паренхимы, клетки стромы, внутриутробное антигенное действие, гликопротеины, крысы.