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Immunohistochemical features of the ovaries' structure in case of fetuses with a gestational term of 21-28 weeks from mother, whose pregnancy was complicated by chronic infections of lower genital tracts

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Abstract

The following article presents results of the complex research on postulation of immunohistochemical features of the ovaries' structure in case of fetuses with a gestational term of 21-28 weeks from mothers with HILGT comparing to ones in case of fetuses from mothers with a physiological pregnancy. The materials of the research are: 25 fetuses from mothers with HILGT as well as 25 fetuses from mothers with physiological pregnancy. All fetuses had died antenatally and inranatally, as a result of violation of uteroplacental and umbilical-uterine circulation. Research methods: macroscopic, organometric, histological, immunohistochemical, morphometric, statistical. The organometric method revealed a clear decrease of main indexes of the fetuses' ovaries from mothers with a complicated pregnancy comparing to ones in case of organs of fetuses from healthy mothers. Morphometric method revealed a clear decrease of a relative volume of cortical substance as well as a clear increase of a relative volume of brain substance in the gonads of fetuses from the main group; as well as it showed a clear decrease of an index of follicular tissue' relative volume, likewise a clear increase of an index of interstitial component relative volume.

The staining by picrofuxin by van Gieson method revealed a massive growth of the connective tissue in all structural components of organs of fetuses from mothers with HILGT relatively to gonads of fetuses from healthy mothers. Using of MCAT to main types of collagens allowed to reveal a violation of maturation of main types of collagens in the structure of connective tissue of fetal gonads in the main group. In the follicular component of the fetuses' ovaries in the main group are appearing mature, cystic-atretic and growing forms of follicles. Immunohistochemical method revealed an increased number of an apoptotically altered eggs in the ovaries of fetuses from mothers with HILGT comparing to one in organs of fetuses from mothers with physiological pregnancy. The peroxidase method revealed an increased endotheline-producing activity of the vascular component as arterial, like a venous type; likewise a clear decrease of the hormone-producing activity in the ovaries of fetuses from mothers with HILGT comparing to such indexes in fetal organs of fetuses from mothers with physiological pregnancy. The complex of changes in the ovaries of fetuses with a 21-28 weeks of gestational term, which was described, was formed under the influence of the chronic hypoxia, prolonged antigenic stimulation as well as hormonal violations in the mother-placenta-fetus system, which is a leading link of the infectious pathology' pathogenesis. The aforementioned complex of the structural and functional changes in the ovaries of fetuses from mothers with HILGT in a gestational term of 21-28 weeks indicates violations in implementation and formation of the fetal gonads, as well as it could further lead to disorders of the germinal function in the female organism.

Key words: ovary; fetus; pregnancy; chronic infection; lower genital tract; collagen; endotheline-1; estrogen.

Імуногістохімічні особливості будови яєчників плодів зі строком гестації 21-28 тижнів від матерів, вагітність у яких ускладнена хронічною інфекцією нижніх статевих шляхів

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В статті наведено результати комплексного дослідження щодо встановлення імуногістохімічних особливостей будови яєчників плодів зі строком гестації 21-28 тижнів від матерів з ХІНСШ порівняно із такими в органах плодів від матерів з фізіологічною вагітністю. Матеріалом дослідження послужили яєчники 25 плодів від матерів з ХІНСШ та 25 плодів від матерів з фізіологічною вагітністю. Всі плоди загинули антенатально та інтранатально внаслідок гострого порушення матково – плацентарного та пуповинно – маткового кровообігу. Методи дослідження: макроскопічний, органометричний, гістологічний, імуногістохімічний, морфометричний, статистичний. Органометричним методом виявлено вірогідне зниження основних показників яєчників плодів від матерів з ускладненою вагітністю порівняно із такими в органах плодів від здорових матерів. Морфометричними методами в гонадах плодів основної групи встановлено вірогідне зниження відносного об'єму коркової речовини та вірогідне підвищення відносного об'єму мозкової речовини, а також вірогідне зниження показника об'єму фолікулярної тканини та вірогідне підвищення показника відносного об'єму інтерстиціального компоненту. Забарвлення пікрофуксином за методом ван Гизон виявило масивне розростання сполучної тканини у всіх структурних компонентах органів плодів від матерів з ХІНСШ відповідно до гонад плодів від здорових матерів. Використання МКАт до основних типів колагенів дозволило встановити порушення дозрівання основних типів колагенів в структурі сполучної тканини фетальних гонад основної групи. В фолікулярному компоненті яєчників плодів основної групи з'являються зріючі, кистозно – атретичні та зростаючі форми фолікулів. Імуногістохімічним методом в яєчниках плодів від матерів з ХІНСШ показано підвищення кількості апоптозно

змінених яйцеклітин порівняно із такою в органах плодів від матерів з фізіологічною вагітністю. Пероксидазним методом встановлено підвищення ендотелін – продукуючої активності судинного компоненту як артеріального, так і венозного типів, а також вірогідне зниження гормон – продукуючої активності в яєчниках плодів від матерів з ХІНСШ порівняно із даними показниками в фетальних органах плодів від матерів з фізіологічною вагітністю. Описаний комплекс змін в яєчниках плодів зі строком гестації 21-28 тижнів сформувався під впливом хронічної гіпоксії, тривалої антигенної стимуляції та гормональних порушень в системі мати – плацента – плід, що є провідними ланками патогенезу інфекційної патології. Наведений комплекс структурних та функціональних змін в яєчниках плодів від матерів з ХІНСШ у термін гестації 21-28 тижнів свідчить про порушення закладки та формування фетальних гонад і може призвести у подальшому до розладів гермінативної функції жіночого організму.

Ключові слова: яєчник; плід; вагітність; хронічна інфекція; нижні статеві шляхи; колаген; ендотелін-1; естроген.

The relevance of the topic is prescribed by a widespread prevalence of an infectious pathology among pregnant, as well as by a significant percentage of perinatal losses due to it [1, 2]. The complications of pregnancy by a chronic infection are dangerous because of the fact, that infectious agents are contributing to development of hypoxia in a placental complex, are stimulating collagen formation in walls of vessels and main components of placenta, as well as are factors, that are triggering apoptotic changes in the tissues of fetus as well as are leading to malformations of varying severity [3, 4]. Moreover, we have to notice, that in the situation of lack of the obligatory screening, this presence of an infectious pathology remains undiagnosed, as a result, woman undergo a necessary medical treatment, so the development of fetus takes place in adverse conditions [5, 6]. According to the statistical data, more, than 90% of pregnant women suffer from infectious pathology [7]. Moreover, in most of cases there is a complete lack of manifestations of presence of the infectious process, so the course is asymptomatic [8]. However, it is known, that actually asymptomatic course is accompanied by significant disorders of embryo- and fetogenesis. There were specific changes described from the side of cardiovascular, digestive and immune systems of fetuses, the development of which is occurring under a condition of infection.

However, the immunohistochemical features of the ovaries' structure of fetuses with a gestational term of 21-28 weeks from mothers with a chronic infection of lower genital tracts have not been studied yet.

The main aim of study: to disclose immunohistochemical features of the ovaries' structure of fetuses with a gestational term of 21-28 weeks from mothers, whose pregnancy is complicated by chronic infection of lower genital tracts.

The study material: ovaries of 25 fetuses from mothers, who suffer from chronic infection of lower genital tracts (HILGT) (main group) comparing with 25 gonads of fetuses from mothers with a physiological pregnancy {the group of comparison). All fetuses had died antenatally and intranatally, as a result of acute violation of uteroplacental circulation (detachment of the normally located placenta) as well as umbilical-fetal circulation (umbilical cord pathology).

The presence of infection in a feto-placental complex in the main group is confirmed in every case by applying modern research methods towards areas of maternal and fetuses' internal organs (by bacteriological, biochemical, as well as by polymerase chain reaction). More and more often the following pathogens were noticed: herpes virus, cytomegalovirus infection, enterococcus, chlamydial infection, *Escherichia coli*, staphylococcus, streptococcus, toxoplasmosis, *Kl. Pneumoniae*, *p. Mirabilis*, *E. cloacae*, *p. Aeruginosa*, as well as the following combinations: cytomegalovirus and chlamydial infections, chlamydial infections and *Escherichia coli*, herpes virus with cytomegalovirus infection.

Women from the group of comparison were healthy according to the medical documentation (medical records of pregnancy development, maps of the research during the stay in a maternity hospital).

Research methods: macroscopic, organometric, histological, immunohistochemical, morphometric, statistical.

After removal all organs were examined, and the main dimensions and indexes were measured. From every organ there were three pieces from different functional zones removed so, that all layers of wall fell into a specimen. The parts of organs were fixed and performed through alcohols of different concentration. Fixed pieces were filled with paraffin, as a result of what blocks were made. From each block there were 3-5 sections made, that were stained by histological and immunohistochemical methods.

Histological methods included staining by hematoxylin and eosin, as well as staining by the method of van Gieson [9, 10].

The study on specimens, that were stained by histological methods, as well as morphometric research had been performed on the Olympus BX-41 microscope using Olympus Db-soft (Version 3:1) software [9, 10, 11].

The immunohistochemical study was held by applying a direct Koons method with a modification of M. Brosman (1979) with using MCAT to collagens of the I, III and IV types, to endotheline-1 and CD 95 (Chemicon International Inc., Temecula California), as well as by peroxidase method [11, 12]. The specimens that had been treated by a direct Koons method were studied on fluorescent “Axioskop 40” (Carl Zeiss, Germany) microscope. The optical density of immunofluorescence of collagens of the I, III and IV types, endothelin-1, as well as a number of apoptotically altered eggs, were identified by applying a method of G.I. Gubina-Vakulik and co-authors (G.I. Gubina-Vakulik, I.V. Sorokina, V.D. Markovskiy, L.S. Kupriianova, R.V. Sydorenko. Method of quantitative determination of antigen content in biological tissues. Patent for utility model № 46489; CO 1 № 33/00 from 25.12.2009, Bulletin № 4). The researches were held on “Axioskop 40” microscope using Biostat.exe. software.

The evaluation of immunohistochemical reactions with treatment of specimens by MCAT to estrogen and progesterone were held by taking into account both the intensity of staining, as well as establishing a percentage of immunopositive cells relatively to a general number of cells [10, 11]. As MCAT an estrogen-receptor alfa (ER), (DAKO clone EP1) and progesterone-receptor (PR), (DAKO clone PgR 636) had been used.

The statistical processing of an obtained data was held on the personal computer using statistical packages „Excell for Windows”, „Statistica 7.0. for Windows”, „SigmaStat 3.1. for Windows” 1 [12, 13].

The results and its discussions. The ovaries of fetuses from the study groups were located horizontally close to the side wall of the pelvis. In all cases the size of the right ovary prevailed towards the left one. The shape of gonads was triangular, flattened. The surface of the ovaries of fetuses from the study groups is hilly, bluish-white. In the section, the tissue of fetal organs is grayish-pink, with a homogeneous consistency.

The organometric indexes of the fetuses' indexes from study groups are presented in the Table 1.

By taking into account data from the Table 1, we could come up with the conclusion, that indexes of weight, width, thickness and volume of the ovaries of the fetuses from mothers with HILGT are clearly decreased comparing with the same indexes in case of healthy mothers.

Table 1 - The organometric indexes of the fetuses' ovaries from study groups

Groups	The ovary weight, kg	The ovary length, m	The ovary width, m	The ovary thickness, m	The ovary volume, m ³
Group of comparison	1,89±0,02 x10 ⁻³	1,85±0,03 x10 ⁻²	1,78±0,06 x10 ⁻²	1,31±0,02 x10 ⁻²	0,039±0,001
Main group	1,75±0,06 x10 ⁻³ *	1,81±0,06 x10 ⁻²	1,64±0,06 x10 ⁻² *	1,15±0,03 x10 ⁻² *	0,034±0,001*

Note p≤0,05.

The ovaries of the fetuses from the both groups are covered with a protein shell, the thickness of which reaches in average the following indexes: in the group of comparison - $24,67 \pm 0,26 \times 10^{-3} \text{ m}$, in the main group - $22,18 \pm 0,81 \times 10^{-3} \text{ m}$. It means, that the thickness of the protein shell in case of ovaries of fetuses from mothers with a physiological pregnancy prevails towards the same index in case of ovaries of fetuses from mothers, whose pregnancy was complicated by HILGT. Histological method revealed a prevalence of thin collagen fibers in the structure of the protein shell of gonads in study groups.

On the current stage of the in utero development the division between cortical and cerebral layers is unclear. However, we can still measure main indexes of relative volumes of the layers. Thus, the average relative volume of the cortical substance reaches $61,2 \pm 2,14 \%$ in case of fetuses from the group of comparison; $57,34 \pm 2,01\%$ - in case of fetuses from the main group; while the relative volume of the cerebral substance reaches $38,8 \pm 1,36 \%$ - in case of fetuses from healthy mothers and $42,66 \pm 1,40\%$ - in case of fetuses from mothers with complicated pregnancy. This data reveals a clear decrease of the relative volume of the cortical substance as well as a clear increase of the relative volume of cerebral substance in gonads of fetuses from mothers with HILGT, comparing to those indexes in case of fetuses from mothers with physiological pregnancy.

The clusters of germ cells in all observations are situated under a protein shell. A great number of cells possesses features of degeneration. Histological methods revealed a decrease of the chromatin content in the nuclei of cells, which indicates their functional activity on this stage of the ovaries' stabilization. Individual cells have pyknotic changes in the nucleus and vacuolation of the cytoplasm. The average number of the germ cells as well as their apoptotically altered forms according to groups are presented in the Table 2.

Table 2 - The indexes of number of germ cells and an apoptotical index in the ovaries of fetuses from the study groups

Group	Number of germ cells, examples in an eyeshot	Apoptotical index, %
Group of comparison	44,89±1,06	22,72±0,80 %
Main group	41,02±1,43*	29,48±1,03*

Note $p \leq 0,05$.

The data from the Table 2 reveals a decrease of number of the germ cells as well as an increase of an apoptotical index in ovaries of fetuses from mothers with HILGT comparing to this index in case of fetuses from mothers with a physiological pregnancy.

The cortical layer of ovaries of fetuses from mothers with a physiological pregnancy is represented by primordial and primary follicles, between which there are layers of the connective tissue. In all gonads we could notice germ cords. Primordial follicles are represented by the oocyte, which is surrounded by squamous epithelial cells and a connective tissue sheath. Primary follicles consist of the oocyte and cells of the follicular epithelium all around it.

In the follicular component of ovaries of fetuses from mother with HILGT there are primary, primordial, maturing, cystic - atretic forms. Growing follicles are situated in the surface layer of the ovary and are characterized by the enlarged egg and cells of the cylindrical epithelium around it. In the maturing follicles epithelial cells are arranged in layers, while there is a cavity in the center filled with eosinophilic fluid. The appearance of maturing, cystic- atretic and growing forms of follicles in the follicula component indicates the acceleration of maturing of fetal gonads, which is not typical for the current stage of in utero development.

The indexes of number of follicles and their diameter are shown in the Table 3.

According to the data from Table 3, in ovaries of fetuses from the main group we could notice a clear increase of number of primordial and primary follicles, as well as an increase of their diameter' index comparing to the same indexes in case of fetuses from the group of comparison. Moreover, in the gonads of fetuses from mothers with HILGT are appearing growing, maturing as well as cystic-atretic forms of follicles, what is typical for the aforementioned stage of in utero development of fetus.

Table 3 - The indexes of numbers of follicles and their diameter in ovaries of fetuses from study groups

Index/Group	Follicles				
	Primordial	Primary	Growing	Maturing	Cystic-atretic
Number of follicles/Group of comparison	31,63±1,11	35,70±0,02	--	--	--
Number of follicles/ Main group	33,07±1,16*	38,19±1,34*	0,78±0,03	0,41±0,01	17,24±0,63
Diameter of follicles/Group of comparison	30,08±1,06 x10 ⁻⁶	32,49±1,14x 10 ⁻⁶	--	--	--
Diameter of follicles/ Main group	33,16±1,16x 10 ⁻⁶ *	35,62±1,22x 10 ⁻⁶ *	78,15±2,74x 10 ⁻⁶	17,69±0,62 x10 ⁻⁶	3,15±0,11x 10 ⁻⁶

Note p≤0,05.

In the ovaries of fetuses from mothers with a physiological pregnancy the collagen of the IV type prevails in the structure of connective tissue in a basement membrane; while the collagen of the III type as appearing as a small focal glow of a low intensity. Thus, the intensity of glow of collagen of the IV type reaches: 1,04±0,04 conventional units of optical density (conv. un. opt. dens.); towards the collagen of the III type - 0,94±0,03 conv. un. opt. dens.). In case of organs of fetuses from mothers with complicated pregnancy the collagen of the III type prevails in the structure of basement membrane, while the indexes of glow of the collagens are reaching as follows: for the collagen of the IV type - 0,88±0,04 conv. un. opt. dens.; for the collagen of the III type - 1,23±0,03 conv. un. opt. dens.. Thus, we can notice, that in case of fetuses from mothers with HILGT, comparing to fetuses from mothers with physiological pregnancy there is a violation of the collagen formation in the follicles' basement membranes.

The aforementioned changes of the collagen formation, that had been found out by the Koons method, are confirmed by peroxidase method. Thus, in the follicles of ovaries of fetuses from the group of comparison it is shown an excessive reaction (+++) while treating the specimens by MCAT to the collagen of the IV type and a moderate reaction (++) while treating specimens by MCAT to the collagen of the III type. In the follicles of organs of

fetuses from the main group it was postulated, that oppositely, there is a moderate reaction (++) while treating specimens by MCAT to the collagen of the IV type as well as an excessive reaction (+++) while treating specimens by MCAT to the collagen of the III type.

By applying picrofuxin by the van Gieson method it was postulated, that in all gonads connective tissue is staining in a red color. Treating the specimens by MCAT to main types of the collagens revealed the following features of the connective tissue structure in ovaries of fetuses from the study groups. Thus, the intensity of glow of the collagen of the I type in a group of comparison reaches $3,78 \pm 0,13$ conv. un. opt. dens., in a main group - $3,16 \pm 0,11$ conv. un. opt. dens.; the intensity of glow of the collagen of the III type in ovaries of fetuses from the group of comparison is - $2,64 \pm 0,09$ conv. un. opt. dens., in organs of fetuses from the main group - $4,31 \pm 0,15$ conv. un. opt. dens. By analyzing the aforementioned indexes we come up with understanding, that in gonads of fetuses from mothers with HILGT there is a violation of the collagen formation. Namely: the collagen of the III type prevails comparing to the collagen of the I type. In ovaries of fetuses from mothers with complicated pregnancy the indexes of intensity of glow of the collagen of the I type are clearly decreased, while those of the collagen of the III type are clearly increased comparing to the same indexes in case of fetuses from mothers with physiological pregnancy.

The aforementioned changes are confirmed by peroxidase method. Thus, in the structure of the connective tissue of fetuses' organs from the group of comparison the following features were revealed: there is prevalence of the collagen of the I type (an intensive reaction - +++) and a moderate reaction while treating specimens by MCAT to the collagen of the III type (mild expression - +). The connective tissue of ovaries of fetuses from the main group is characterized by an intensive reaction (++) to the collagen of the III type as well as by a moderate reaction while treating by MCAT to the collagen of the I type (++).

The average indexes of the relative volumes of main structural components in the ovaries of fetuses from the study groups are presented in the Table 4.

Table 4 - The indexes of volumes of main structural components of fetuses' ovaries from the study groups, (%)

Group	Interstitial tissue	Follicular tissue
Group of comparison	$26,4 \pm 0,61$	$73,6 \pm 2,54$
Main group	$29,60 \pm 1,04^*$	$70,42 \pm 1,41^*$

Note $p \leq 0,05$.

Thus, in the organs of fetuses from the main group we notice a clearly increased relative volume of the interstitial tissue as well as clearly decreased relative volume of the follicular tissue comparing to the same indexes in case of organs of fetuses from the group of comparison.

The vascular component in all observations is represented by arteries and venous of a moderate blood supply. The plane indicators of vessels of gonads of the fetuses from mothers with a physiological pregnancy are reaching in general: in a cerebral substance: $35,11 \pm 1,23$ %, in a cortical one - $16,72 \pm 0,59$ %; in ovaries of fetuses from mothers with a complicated pregnancy: in a cerebral substance the index reaches: $33,16 \pm 1,16$ %, in a cortical layer - $14,25 \pm 0,5$. The data, which was provided before, indicates a clear decrease of indexes of the vessels' plane as in the cerebral substance, as in the cortical layer of ovaries of fetuses from mothers with HILGT relatively to the same data in case of organs of fetuses from healthy mothers.

Immunohistochemical method revealed an optical density of the immunofluorescence of endothelial cells in vessels of the ovaries of fetuses from the group of comparison, the indexes of which are reaching as follows: $0,32 \pm 0,01$ conv. un. opt. dens. - in the vessels of the arterial type; and $0,64 \pm 0,02$ conv. un. opt. dens. - in the vessels of the venous type. In the vessels of organs of fetuses from the main groups we can see as follows: in the vessels of the arterial type - $0,81 \pm 0,03$ conv. un. opt. dens., and in the vessels of the venous type - $1,24 \pm 0,04$ conv. un. opt. dens. Thus, by analyzing the aforementioned indexes we can come up with the following conclusion: in the vessels both of arterial and venous types in ovaries of fetuses from mothers with HILGT there is a highly increased level of the endotheline-producing activity relatively to ones in case of organs of fetuses from mothers with physiological pregnancy.

Applying of MCAT to the ovaries' hormones revealed the following features in the gonads of the study groups. Thus, in case of fetuses from mothers with a physiological pregnancy there are an excessive reaction for estrogen (+++, up to 80% of cells) and a negative one to progesterone (- -). In case of organs of fetuses from mothers with HILGT there are a moderate positive reaction for estrogen (++) and a negative for progesterone. During the quantitative evaluation it was postulated, that in case of 65% of cells, on the surface of which there are estrogen receptors, we could notice a moderate positive reaction while treating by MCAT to estrogen. By comparing these indexes we can come up with the following conclusion: the hormonal activity of organs of fetuses from the main group is decreased relatively to one in case of organs of fetuses from the group of comparison.

Thus, in the ovaries of fetuses from mothers, whose pregnancy was complicated by HILGT, there were revealed immunohistochemical features, which are characteristic for violation of implementation and maturing of the organ. The organometric indexes of fetuses' organs in the main group are clearly decreased comparing to ones in case of group of comparison, what is determined by vascular disorders in placenta [14, 15]. The relative volume of the cortical substance is decreased, while the same index of the cerebral one is increased as a result, first of all, of increasing of indexes of the follicles' number and their diameter, as well as because of the decrease of number of the germ cells. The postulated changes are caused primarily by an action of the chronic hypoxia and infectious agent, as well as hormonal dysfunction of placenta, the development of which is typical under a condition of this pathology of pregnancy [16, 17]. Immunohistochemical method revealed an increase of the apoptotical index among germ cells, what is stimulated by an action of the infectious agent during maturing of the organism' cells [18, 19]. In gonads of fetuses from mothers with HILGT are appearing maturing, cystic-atretic and growing follicles, what could be a feature of the functional overstrain of organ during a period of in utero development [20]. Histological methods revealed a decrease of indexes of vascular areas as in the cerebral, as in the cortical substance, what is caused by hyperplasia of placenta under the condition of the chronic infection [21].

The chronic hypoxia, under the condition of which fetuses are forming, contributes to the increased collagen formation in all structural components of ovaries, as well as lead to violation of maturing of the main collagens' types [18, 20].

In the vessels both of arterial and venous types in case of gonads of fetuses from mothers with HILGT we could notice an increase of the endotheline-producing activity comparing to vessels in ovaries of fetuses from mothers with a physiological pregnancy. It is caused by chronic hypoxia, which is formed under the influence of infectious agents as well as violations in the vascular component in the mother-placenta-fetus system [17, 19].

Peroxidase method revealed a decreased number of cells, which are expressing receptors to estrogen, in organs of fetuses in the main group. The decrease of hormonal activity in ovaries of fetuses is caused by a violation of implementation and formation of main structural components of the fetal gonads.

The postulated changes in the ovaries of fetuses from mothers with HILGT comparing to ones in case of organs of fetuses from mothers with physiological pregnancy are caused by gross violations in placenta as well as by changes in the mother's organism [17, 19].

Conclusions

1. Organometric indexes of the ovaries (weight, length, thickness, width and volume) of fetuses from mothers with HILGT are clearly decreased relatively to these indexes in case of fetuses from mothers with physiological pregnancy. Unilaterally directed changes are observed during assessment of indexes of the protein shell' thickness of the fetal gonads.

2. Morphometric indexes of the relative volumes of cortical and cerebral substances in the structure of fetal organs had been changing as follows: in ovaries of fetuses from mothers with a complicated pregnancy the relative volume of the cortical substance is clearly decreased, while the relative volume of the cerebral substance is clearly increased comparing to the same indexes in case of ovaries of fetuses from mothers with physiological pregnancy.

3. By applying histological methods it was shown a decrease of number of the germ cells in fetal gonads of fetuses from the main group comparing to this number in ovaries of fetuses from the group of comparison.

4. Immunohistochemical method revealed an increase number of the apoptotically altered eggs in ovaries of fetuses from mothers with complicated pregnancy comparing to this index in case of organs of fetuses from mothers, whose pregnancy was physiological one.

5. Follicular component of ovaries of fetuses from the main group differs from one in case of organs of fetuses in the group of comparison by appearing cystic-atretic, growing and maturing forms of follicles, as well as by growing number of primary and primordial follicles as well as their diameter parameters.

6. Morphometric method revealed a clear decrease of indexes of the relative volume of follicular tissue as well as a clear increase of indexes of the relative volume of interstitial component in gonads of fetuses from mothers with HILGT comparing to these parameters in case of ovaries of fetuses from mothers with a physiological pregnancy.

7. Histological methods have shown a massive growth of the connective tissue in the cerebral and cortical substances of organs of fetuses from the main group comparing to ovaries from the group of comparison. Immunohistochemical methods have postulated a violation of maturing of main collagens'types in the structure of connective tissue of the structural components of organs of fetuses from mothers with a complicated pregnancy relatively to the structure of one in ovaries of fetuses from mothers with physiological pregnancy. Moreover, there is a shown increased glow of the collagen of the III type as well

as a decreased intensity of glow of the collagen of the I type and IV type in the structure of the connective tissue as well as vascular wall in ovaries of fetuses from mothers with HILGT.

8. Morphometric method has postulated a decrease of indexes of the plane of vessels in ovaries of fetuses from the main group relatively to one in organs of fetuses from the group of comparison.

9. Immunohistochemical method has revealed a clear invrease of glow of the endotheline-1 in vessels both of arterial and venous types of ovaries of fetuses from mothers with complicated pregnancy relatively to these indexes in case of organs of fetuses from healthy mothers.

10. In the ovaries of fetuses from mothers with a complicated pregnancy we have noticed a decrease of the hormone-producing activity relatively to one in organs of fetuses from mothers with a physiological pregnancy.

11. The aforementioned changes in ovaries of fetuses from the main group (namely: decrease of organometric indexes, massive growth of the connective tissue in the strucutral components of organ, violation of maturing of the main collagens' types, an increase o the endotheline-producing activity of the vascular component) are caused by a chronic hypoxia in the mother-placenta-fetus system, which is a leading link under the conditions of infectioning.

12. The appearance of maturing, cystic-atretic and growing forms of follicles in the follicular component of ovaries of fetuses from mothers with HILGT is caused by a chronic hypoxia as well as by constant antigenic stimulation under the conditions of this complication of pregnancy as well as they lead to accelerated maturing of the fetal gonads.

13. The immunohistochemical features of the ovaries' structure in case of fetuses from mothers with a complicated pregnancy, that had been postulated, are determing gross violations of implementation and formation of ovaries of fetuses on the current stage of in utero development, likewise they could further lead to formation of organ failure in the subsequent onotgenesis.

Perspectives of the subsequent reseach: to disclose immunohistochemical features of the ovaries' structure in case of fetuses from mothers, whose pregnancy was complicated by chronic infection of the lower genital tract, with gestational terms of 29-36 weeks as well as 37-40 weeks.

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