



Ontogenesis of the erythroblastic islands in swine

^{1,2}Marina Tatoyan, ²Sergey Karapetyan, ²Lina Hakobyan, ²Liana Abroyan, ²Aida Avetisyan, ²Elena Karalova, ²Zaven Karalyan

¹ Yerevan State Medical University, M. Heratsi Department of Histology, Cytology and Embryology; ² Laboratory of Cell Biology, Institute of Molecular Biology of NAS RA.
Corresponding author: M. Tatoyan, marina.tatoyan@mail.ru

Abstract. In the 15-days pig embryo EI with the central macrophage is not available. Initially the morphologically similar formations (without a phagocytated nucleus) are coming apparent just in the 25-days pig embryo and the full-fledged hepatic EI formation with the central macrophage and the phagocytated nuclei of erythroblasts refers approximately to the 55th day of the pig embryonal development. An extrusion of the erythroblast's nucleus with its following phagocytosis by the central macrophage of island, what is highly typical for erythroblastic islands, was also shown. Thus, two types of erythropoiesis – the primitive and definitive ones are gradually interchanged in the liver during the coming to being process of the pig embryo's hemopoiesis.

Key Words: macrophage, erythropoiesis, erythroblast, embryonal development.

Абстракт. При изучении эмбрионального эритропоеза у 15 суточного эмбриона свиней в эритробластических островках с центральным макрофагом не выявлено. Первые морфологически сходные образования (без фагоцитированного ядра) появляются у 25 суточного эмбриона. А формирование полноценных печеночных эритробластических островков с центральным макрофагом и фагоцитированными ядрами эритробластов относится примерно к 55 суткам развития плода свиньи. Также нами показана характерная для эритробластических островков экструзия ядра эритробласта с ее последующим фагоцитозом центральным макрофагом островка. Таким образом, в печени последовательно сменяются два типа эритропоеза – примитивный и дефинитивный.

Ключевые слова: макрофаги, эритропоез, эмбриональное развитие, эритробласты.

Introduction. Erythroblastic island (EI) is a distinct histological unit which consists of a central macrophage surrounded by the erythroid cells. These structures are present in the fetal liver and the adults bone marrow, and have a key role in erythroid cells proliferation and differentiation (Mao et al 2013).

Hemopoiesis in the embryonal development of mammals is started with the yolk sack and then is being transferred into the liver. Hemopoiesis in the liver is occurred in extra-vascular way, nevertheless taking place along the vessels main direction. The stem hematopoietic cells are differentiated into the blasts, and thereafter into the secondary erythrocytes. Development of erythroblastic islands with a central macrophage in it is taking place in the liver. In pigs the hemopoiesis is transferred into the liver starting from the 20th day of embryo's development, after which the liver becomes the main site of hemopoiesis and stands active till first weeks after the birth, but with the activities gradual attenuation. But already at the early stages of embryos development in the mammals vessels the nucleus-containing cells of much lesser size as compared to the primitive hematopoietic cells are observed. To that period of time the nonnuclear erythrocytes are formed, which are of lesser size comparably to the cells, arising in the yolk sack hematopoietic islands. It testifies to different types of erythropoiesis availability at the mentioned period of the pig embryo's development (Isern & Méndez-Ferrer 2011; Tatoyan et al 2015). As it is known, a loss of nuclei is a unique behavior during definitive type of erythroid maturation in mammals (Dessypris 1993), and the extruded nuclei are immediately taken up by macrophages of EI. These erythroid cells belong to new

generation of cells (hepatic proper cells) and are capable of maturation in the erythroblastic islands structure.

According to the contemporary conception macrophages are of crucial importance in regulation of the hematopoietic cells functional activity and namely by these macrophages of EI the specific micro-surrounding is being formed, providing the erythroid cells proliferation and differentiation (Giger & Kalfa 2015).

The pig embryo's hemopoiesis is still less studied today (Pearson et al 1998; Vallet et al 2003), but the EI are not investigated at all.

The aim of the present study is to bring a more clear light on porcine erythroblastic islands in ontogenesis.

Material and Method

Animals. 16 sows, Large White breed, aged in 11 months were used in the given experiment. The pregnant sows being grown up till 130-140 kg in 11-12 months of age underwent slaughter to study the embryos and fetuses development and growth.

A slaughter was done on the 15th, 25th, 35th, 45th, 55th, 65th, 75th and 90th days of the sows pregnancy. For each mentioned day two sows were slaughtered. Also the piglets of 24 hours, 30 days and 90 days of birth were studied. For each time not less than 8 embryos (fetuses) and 3-5 piglets were studied. Euthanasia of the pigs was performed according to the Guide for Care and Use of Laboratory Animals, AVMA Guidelines (International Review Board/Independent Ethics Committee of the Molecular Biology of NAS, IRB00004079).

Laboratory investigation. As fixations for the histological samples of embryos the Zenker, Flemming, Buen liquids and 96% ethanol solution were used. The samples were flooded in paraffin, followed by preparations of the series of histological slides of 5-8 μm thick. The preparations were stained by Hematoxylin solution according to Weigert (Suvarna 2012) and then by eosin (Suvarna 2012) and azan according to Mallory's trichrome (Gray 1954).

Morphometric analysis of the liver. The embryo's liver hematogenic tissue size was determined on the preparations slides with the help of programming support ImageJ. In each case not less than 4 liver slides were used, and in each slide not less than 30 fields of view (0.4 mm) were observed. Fields of view have been chosen randomly, excluding the liver capsule.

DNA protein staining. All preparations were treated with the combined Feulgen-Naphthol Yellow staining (FNYS) procedure (Lillie 1965; Gaub et al 1975). This method permits simultaneous microspectrophotometric analyses of DNA and protein in single cells and the protein value is closely correlated to the amount of dry mass of the cell.

Image scanning cytometry. For image scanning cytometry and DNA measurement, blood slides were fixed in 96% ethanol for 30 minutes and stained in fresh Schiff reagent (DNA hydrolysis in 5 N hydrochloric acid for 60 minute at 22°C) by the method of Deich (1966). In order to measure DNA content (in conventional units) by image scanning cytometry, computer-equipped microscope-cytometer SMP 05 (OPTON) was used at 575 nm wave length and at 1250 \times magnification. Before the scanning process, each nucleus was contoured, and cytometry of nuclear DNA content of all studied types of cells were carried out at 1 to 7 dpi.

Ploidy of cells. DNA content was expressed on a "c" scale, in which 1 c is the haploid amount of nuclear DNA occurred in normal (non-pathologic) diploid populations in G0/G1. The DNA content of unstimulated swine lymphocytes was used as a diploid standard for measurements. DNA measurements identify nuclei as aneuploid if they deviate more than 10% from 2 c, 4 c, 8 c, or 16 c; i.e. if they are outside of $2c \pm 0.2$, $4c \pm 0.4$, $8c \pm 0.8$, or $16c \pm 1.6$ values. The total number of cells in euploid areas of the DNA histogram rescaled

by the mean corrective factor (1.8c-2.2c, 3.6c-4.4c, 7.2c-8.8c, and 14.4c-17.6c) was also calculated. The variability of DNA content in unstimulated lymphocytes did not exceed 10%.

Statistical analysis. The significance was evaluated by two-tailed Student's t-test.

Results. Starting approximately from the 20th day of pig embryos development the hemopoiesis is transferred into the liver, which then gradually becomes the major active site of hemopoiesis, although afterwards its leading role in erythropoiesis is attenuated till the birth moment. It should be noted also, that at the initial stage of hepatic hemopoiesis a formation of the primitive hematopoietic cells, analogical to those in the yolk sac is encountered (Isern & Méndez-Ferrer 2011), but the erythroblastic islands are not available (Figure 1). Primitive hematopoietic cells very soon form the hemopoiesis loci, alike the yolk sac blood islands (Figure 2.). Anyway, even at the early stages of hemopoiesis (the 25th days of the embryo's development) formation of the cells of a lesser comparably to primitive hematopoietic cells size (Figure 1.) is observed. It testifies to an onset of the normoblastic hemopoiesis.

The mature non-nucleated erythrocytes (of a lesser size comparably to the primitive ones) are revealed in the embryos vessels already at the age of 25 days, but in a significant number they are occurred in the circulation much later. From the pigs erythropoiesis point of view the 25-days embryo is of great interest. Along with the primitive erythropoiesis taking place in the pig liver starting with the 25th day of development the cells of a lesser size, the mature forms of hematopoietic cells, are also revealed.

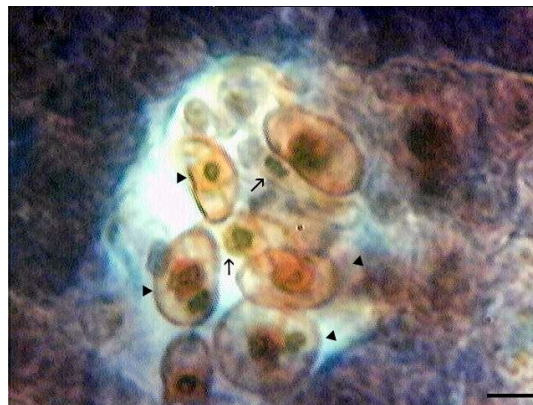


Figure 1. Longitudinal section the 25 days aged liver. Presence of two types of erythropoiesis. Staining: with trichrome by Mallory. Primitive hematopoietic cells – megaloblastic erythropoiesis (red colored, shown by black triangles) 25 days swine embryo. Normoblastic erythropoiesis (erythroblasts are shown by black arrows). Scale bar is 10 μ m (original).

Thus, at this stage of embryos development two different systems of erythropoietic cells (the primitive and erythroblastic ones) are circulated in its vascular system. Each one of these systems of cells is represented by several types of cells (Tatoyan et al 2015).

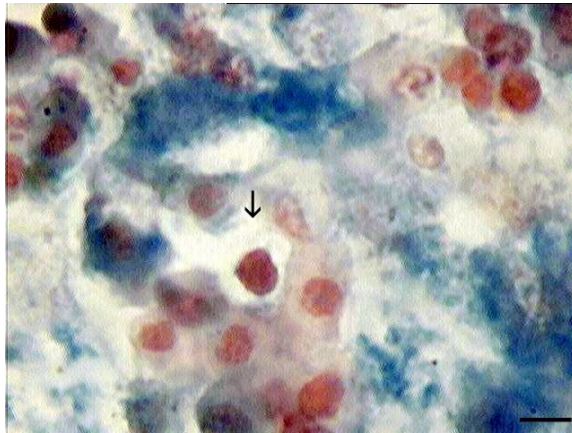


Figure 2. Longitudinal section of the 25- day liver. Staining: by Giemsa. The macrophage –alike cell (is shown by an arrow) in the hematopoietic cells surrounding. Scale bar is 10 μ m (original).

At this period of time the cells morphologically different from hematopoietic ones are originated in the liver. These are huge cells of irregular form having a great share of cytoplasm and a slightly staining capacity by Giemsa. The nuclei of these cells comparably to the hemopoietic ones are stained in more intensive way. In analysis of the nuclei distribution according to the ploidity classes all the cells of the given period of development were diploid.

Starting from the 45th day of embryos development a significant number of erythroblastic islands in the liver with the central macrophage in them is revealed (Figure 3 A, B). An extrusion of the erythroblast's nucleus with its following phagocytosis by the central macrophage of island, what is highly typical for erythroblastic islands, was also shown by us. Thus, two types of erythropoiesis – the primitive and definitive ones are gradually interchanged in the liver during the coming to being process of the pig embryos hemopoiesis.

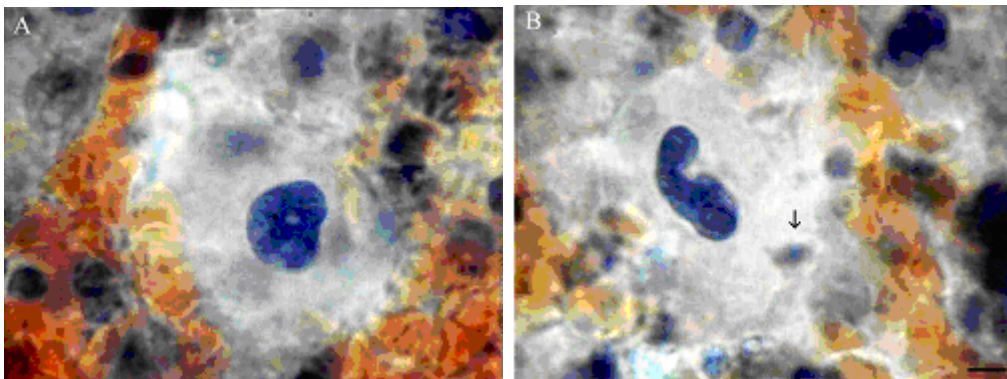


Figure 3. Longitudinal section of the 55-days pig fetus liver. Staining: by hematoxylin-eosin, and additionally with sudan III. An erythroblastic island. The phagocytated nucleus of erythroblast is visible (it is shown by an arrow). Scale bar is 10 μ m (original).

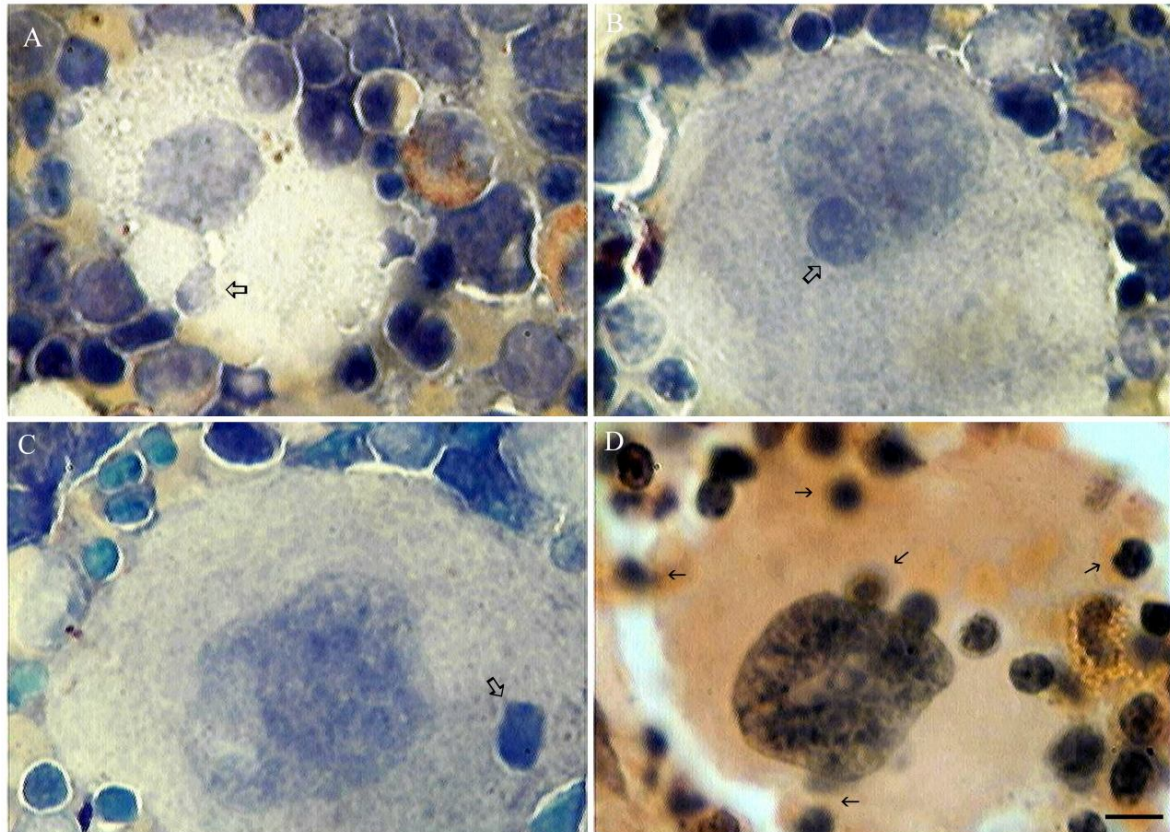


Figure 4. Bone marrow EI of 3 months aged piglets. A. B. C. EIs in the bone marrow aspirates. Staining by Giemsa. Extruded nuclei (transparent arrows); D. EI in a section of the bone marrow. Staining by hematoxylin-eosin. Erythroblasts (black arrows); Phagocytated nuclei (transparent arrows). Scale bar is 10 μ m (original).

As followed from Figure 2, macrophages of the liver EI showed active phagocytosis, and the cytoplasm is occupied by phagocytated nuclei extruded from erythroblasts of the definitive erythropoiesis.

After the first month of gestation erythropoiesis is gradually moving in the bone marrow. And bone marrow erythropoiesis becomes the prevailing at the end of 3th month of gestation. In bone marrow there is only erythropoiesis in erythroblastic islands (Figure 4).

As followed from the Figure 5, the main population of the hepatic EI macrophages is diploid and only insignificant moiety of them (on the 55th day of fetus development) is heteroploid. To the 75 days of embryonic development liver EI again become diploid. For macrophages of the bone marrow EIs a presence of both the diploid and heteroploid, and also the tetra- and hypertetraploids cells is characteristic. The DNA content in the central macrophages nuclei of the bone marrow EI is getting increased along with the age increasing. If in newborn piglets more than 80% of the bone marrow macrophages are diploid, then in the 3-monthed ones the diploid macrophages compose about 50% of the bone marrow macrophages whole number.

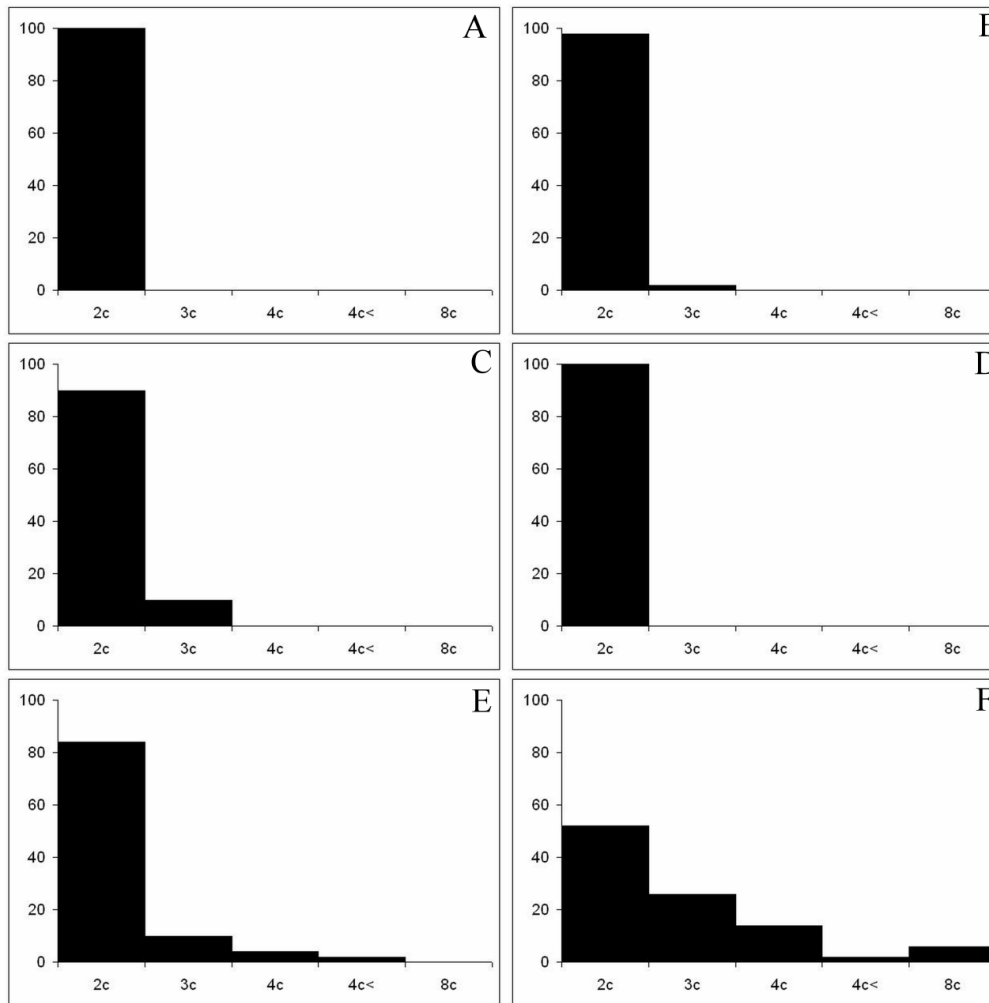


Figure 5. Distribution of nuclei of the EI central macrophages by ploidy. A. 45 days liver EI macrophages; B. 55 days liver EI macrophages, C. 65 days liver EI macrophages, D. 75 days liver EI macrophages, E. Newborn BM EI macrophages, F. 3 months aged piglet BM EI macrophages. X axis - is the ploidy of cells. Y axis - is the percent of the cells distribution.

As it is apparent from the Table 1, the square area of the mature EI in bone marrow predominates over that of the hepatic EI more than two times. Herein, in surrounding of the bone marrow EI central macrophage there are two times more of the nucleated erythroid cells when compared to the hepatic ones.

The proteins contents in the investigated EIs were not of authentic differences.

Table 1
Cytometry and functional indices of the porcine erythroblastic islands in the liver and bone marrow

<i>Localization</i>	<i>Macrophage area (μm^2)</i>	<i>Macrophage protein (cu)</i>	<i>Number of nucleated erythroid cells in EI</i>
Liver EI	702.2±69.7	188.1±53	4.4±1.1
Bone marrow EI, 3 month	1613.5±352*	271.4±68	9.0±1.9*

* significant compared with liver EI ($p < 0.05$).

Discussion. At the early stage of embryo's development (the 15 days embryo) in the liver some discrete loci of hemopoiesis are present. This hemopoiesis is mesoblastic and

identical to the yolk sac hemopoiesis. In this period of development erythropoiesis is taking place in the hematic islands without participation of macrophages (Ferkowicz & Yoder 2005). By the 25th day of pig embryos development in its vessels two different types of cells – megaloblasts (origin is the yolk sac) and normoblasts (origin is the liver) are formed out.

Appearance of the non-nucleated embryonal erythrocytes in the up-dated literature often is connected up with the hepatic EI formation and functioning beginning. Namely in them the erythroblast's nucleus extrusion is occurred (Sonoda et al 1998).

Here is an arising of one of important problems of the embryonal development erythropoiesis, namely determination of the EI origination period. This problem is connected with the erythropoiesis peculiarities in mammals. The erythrocytes of mammals are non-nucleated and undergo definitive maturation by means of the nucleus deprivation (enucleation). In the mature mammals this process is immediately connected with an emergence of EI with the central macrophage in it. At the early stages of erythropoiesis, provided by the cells of yolk sac origin the overwhelming majority of the erythroid cells possess nuclei (Palis et al 2010). Anyway, a part of the primitive erythroid cells are already capable to undergo enucleation and produce non-nucleated megalocytes (Bethlenfalvay & Block 1970; Steiner & Vogel 1973). However, in mammals (actually in the investigated species) the EI formation is encountered in a rather late period of gestation. For example, in mice a formation of EI corresponds approximately to the middle part of gestation period (Sasaki & Sonoda 2000). Study of the embryonal erythropoiesis in pigs revealed a presence of non-nucleated primitive erythrocytes at the early stages of embryogenesis – on the 15th and 25th days of development (Tatoyan et al 2015). Our data point the fact, that EI with the central macrophage is not available in the 15-days pig embryo. Initially the morphologically similar formations (without a phagocytated nucleus) are coming apparent just in the 25-days pig embryo and the full-fledged hepatic EI formation with the central macrophage and the phagocytated nuclei of erythroblasts refers approximately to the 55th day of the pig embryonal development.

Several lines of evidence suggest that macrophages are not essential for enucleation of definitive erythroblasts *in vivo* (Spike et al 2007). In contrast with McGrath et al (2008), Isern & Méndez-Ferrer (2011) have not found enhancement of the primitive erythroid cells enucleation during coculture on the fetal liver macrophages. Rhodes et al (2008) reported that proliferation of definitive erythroblasts is stimulated by coculture with macrophages. So, a possible role for macrophages in proliferation and/or the later steps proceeding in the primitive erythroid cells maturation remains to be evaluated.

We have not revealed any associations of the primitive erythroid cells with the hepatic macrophages in the 25-days pig embryos, but the non-nucleated primitive erythrocytes were available (in insignificant amount).

Thus, a subject regarding to the primitive erythroid cells enucleation (the nucleus decomposition or extrusion with the help of a macrophage) in pigs is still an issue to be revealed.

There is a variation in number of erythroblasts per island in different species. Rat femur sections reveal about 10 cells per island (Yokoyama et al 2002), whereas islands harvested from the human bone marrow contain 5–30 erythroblasts per island (Lee et al 1988).

In the accessible for us literature there are no data referring to the erythroblasts content in EI of pigs. Our data indicate to the presence of generally 7-11 erythroblasts per EI in the bone marrow in the 3-4 month aged piglets.

As followed from the Figure 4, the DNA synthesis is characteristic for the definite part of macrophages of the hepatic EI, which results in the DNA contents increase in nucleus. The given processes are specifically expressed on the 55-56 days of gestation. After the birth of pigs the tetra-, hypertetra- and even octaploid populations of the EI macrophages in the pig bone marrow are becoming apparent.

Considering the data obtained on the liver tissue involvement into erythropoiesis (Figure 5), we could suggest that the DNA content's increase in the hepatic EI macrophages nuclei is connected with the liver participation's input enhancement in the

process of general erythropoiesis, and the DNA content decrease in them is correlated with the hepatic erythropoiesis quenching tendency. Appearance of the poliploid macrophages in the bone marrow EI is predetermined by the EI functional activity level. Probably, the DNA content increase in the macrophages nuclei results in the functional activity enhancement of the whole EI. For the above-mentioned statement's benefit the data on macrophages of the lymph nodes and the spleen (Karalova et al 1990), as well as the similar processes taking place in megacaryocytes and in a number of other cells are the witnesses of it (Anatskaya & Vinogradov 2010; Raslova et al 2003). Thus, the central macrophages nuclei poliploidization phenomenon in the pig bone marrow EI much likely is connected with the functional loading, represented by the erythrocytes increased production.

Conclusions. The DNA content in the central macrophages nuclei of the liver and bone marrow EI is getting increased along with the age increasing, this phenomenon connected with the functional loading, represented by the erythrocytes increased production.

References

- Anatskaya O. V., Vinogradov A. E., 2010 Somatic polyploidy promotes cell function under stress and energy depletion: evidence from tissue-specific mammal transcriptome. *Funct Integr Genomics* 10(4):433-46.
- Bethlenfalvay N. C., Block M., 1970 Fetal erythropoiesis. Maturation in megaloblastic (yolk sac) erythropoiesis in the C57Bl/6J mouse. *Acta Haematol* 44:240-245.
- Deich A. D., 1966 Introduction to quantitative cytochemistry. Academic Press, New York /London, pp. 65-67.
- Dessypris E. N., 1993 Erythropoiesis. In: Wintrobe's clinical hematology. Lee G. R., Bithell T. C., Foerster J., Atens J. W., Lukens J. N. (eds), 9th ed., pp. 134–157, Lea and Febiger, Philadelphia.
- Ferkowicz M. J., Yoder M. C., 2005 Blood island formation: longstanding observations and modern interpretations. *Exp Hematol* 33(9):1041-1047.
- Gaub J., Auer G., Zetterberg A., 1975 Quantitative cytochemical aspects of a combined feulgen-naphthol yellow S staining procedure for the simultaneous determination of nuclear and cytoplasmic proteins and DNA in mammalian cells. *Exp Cell Res* 92:323-332.
- Giger K. M., Kalfa T. A., 2015 Phylogenetic and ontogenetic view of erythroblastic islands. *Biomed Res Int*, doi: 10.1155/2015/873628.
- Gray P., 1954 The microtome's formulary and guide. Blakiston, New York.
- Isern J., Méndez-Ferrer S., 2011 Stem cell interactions in a bone marrow niche. *Curr Osteoporos Rep* 9(4):210-218.
- Karalova E. M., Bakhshinian M. Z., Magakian I. A., 1990 DNA synthesis and content in the macrophage nuclei of normal mice, during carcinogenesis and during the administration of retinoids to the animals. *Tsitologija* 32(1):47-53.
- Lee S. H., Crocker P. R., Westaby S., Key N., Mason D. Y., Gordon S., Weatherall D. J., 1988 Isolation and immunocytochemical characterization of human bone marrow stromal macrophages in hemopoietic clusters. *J Exp Med* 168:1193–1198.
- Lillie R. D., 1965 Histopathologic technical and practical histochemistry. 3rd edition, McGraw-Hill, New York.
- Mao X., Shi X., Liu F., Li G., Hu L., 2013 Evaluation of erythroblast macrophage protein related to erythroblastic islands in patients with hematopoietic stem cell transplantation. *Eur J Med Res* 18:9. doi: 10.1186/2047-783X-18-9.
- McGrath K. E., Kingsley P. D., Koniski A. D., Porter R. L., Bushnell T. P., Palis J., 2008 Enucleation of primitive erythroid cells generates a transient population of "pyrenocytes" in the mammalian fetus. *Blood* 111(4):2409-2417.
- Palis J., Malik J., McGrath K. E., Kingsley P. D., 2010 Primitive erythropoiesis in the mammalian embryo. *Int J Dev Biol* 54(6-7): 1011-1018.

- Pearson P. L., Klemcke H. G., Christenson R. K., Vallet J. L., 1998 Uterine environment and breed effects on erythropoiesis and liver protein secretion in late embryonic and early fetal swine. *Biol Reprod* 58(4):911-918.
- Raslova H., Roy L., Vourc'h C., Le Couedic J. P., Brison O., Metivier D., Feunteun J., Kroemer G., Debili N., Vainchenker W., 2003 Megakaryocyte polyploidization is associated with a functional gene amplification. *Blood* 101(2):541-544.
- Rhodes K. E., Gekas C., Wang Y., Lux C. T., Francis C. S., Chan D. N., Conway S., Orkin S. H., Yoder M. C., Mikkola H. K., 2008 The emergence of hematopoietic stem cells is initiated in the placental vasculature in the absence of circulation. *Cell Stem Cell* 2(3):252-263.
- Sasaki K., Sonoda Y., 2000 Histometrical and three-dimensional analyses of liver hematopoiesis in the mouse embryo. *Arch Histol Cytol* 63(2):137-146.
- Sonoda Y., Sasaki K., Suda M., Itano C., Iwatsuki H., 1998 Effects of colchicine on the enucleation of erythroid cells and macrophages in the liver of mouse embryos: ultrastructural and three-dimensional studies. *Anat Rec* 251(3):290-296.
- Spike B. T., Dibling B. C., Macleod K. F., 2007 Hypoxic stress underlies defects in erythroblast islands in the Rb-null mouse. *Blood* 110(6):2173-2181.
- Steiner R., Vogel H., 1973 On the kinetics of erythroid cell differentiation in fetal mice: I. Microspectrophotometric determination of the hemoglobin content in erythroid cells during gestation. *J Cell Physiol* 81:323-338.
- Suvarna K. S., 2012 Bancroft's theory and practice of histological techniques. 7th edition, Churchill Livingstone, 654 pp.
- Tatoyan M., Karalova E., Hakobyan L., Abroyan L., Avetisyan A., Karalyan N., Karalyan Z., 2015 Ontogenesis of the pig erythroid cells. *Porc Res* 5(1):12-22.
- Vallet J. L., Klemcke H. G., Christenson R. K., 2003 Interrelationships among conceptus size, uterine protein secretion, fetal erythropoiesis, and uterine capacity. *J Anim Sci* 81(9):2352-2356.
- Yokoyama T., Kitagawa H., Takeuchi T., Tsukahara S., Kannan Y., 2002 No apoptotic cell death of erythroid cells of erythroblastic islands in bone marrow of healthy rats. *J Vet Med Sci* 64:913-919.
- *** International Review Board/Independent Ethics Committee of the Molecular Biology of NAS, IRB00004079.

Received: 03 February 2016. Accepted: 17 March 2016. Published online: 21 March 2016.

Authors:

Marina Tatoyan, Yerevan State Medical University, M. Heratsi Department of Histology, Cytology and Embryology, Armenia, Yerevan, 0025, Koryun St. 2; Institute of Molecular Biology of NAS RA, Laboratory of Cell Biology, Armenia, Yerevan, 0014, Hasratyan St. 7, e-mail: marina.tatoyan@mail.ru

Sergey Karapetyan, Institute of Molecular Biology of NAS RA, Laboratory of Cell Biology, Armenia, Yerevan, 0014, Hasratyan St. 7, e-mail: hatussili@yahoo.com

Lina Hakobyan, Institute of Molecular Biology of NAS RA, Laboratory of Cell Biology, Armenia, Yerevan, 0014, Hasratyan St. 7, e-mail: naira_karalyan@yahoo.com

Liana Abroyan, Institute of Molecular Biology of NAS RA, Laboratory of Cell Biology, Armenia, Yerevan, 0014, Hasratyan St. 7, e-mail: z_karalyan@mb.sci.am

Aida Avetisyan, Institute of Molecular Biology of NAS RA, Laboratory of Cell Biology, Armenia, Yerevan, 0014, Hasratyan St. 7, e-mail: aida.avetisyan@inbox.ru

Elena Karalova, Institute of Molecular Biology of NAS RA, Laboratory of Cell Biology, Armenia, Yerevan, 0014, Hasratyan St. 7, e-mail: karalovae@gmail.com

Zaven Karalyan, Institute of Molecular Biology of NAS RA, Laboratory of Cell Biology, Armenia, Yerevan, 0014, Hasratyan St. 7, e-mail: zkaralyan@yahoo.com

This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution and reproduction in any medium, provided the original author and source are credited.

How to cite this article:

Tatoyan M., Karapetyan S., Hakobyan L., Abroyan L., Avetisyan A., Karalova E., Karalyan Z., 2016 Ontogenesis of the erythroblastic islands in swine. *Porc Res* 6(1):1-9.