41.9%, and the frequency of the aforementioned allele amongst the control group was 34.5%.

Conclusion: The results of this study show that there is no statistically significant correlation between MTHFR C677T polymorphism in women with infertility of unknown cause, who are undergoing in vitro fertilization preparation, but also underline the need for further research.

Acknowledgements: Mentor: Professor Dr. Ivana Novaković.

http://dx.doi.org/10.1016/j.pbj.2017.07.094

PS114

Assessing the oxidative modification of proteins in inflamed placenta combined with iron deficiency anemia in the pregnant through histochemical method with bromophenol blue based on Mikel Calvo

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Aim: To set features of OMB in the cytoplasm of decidua cells in basal plate of the placenta at chorioamnionitis with iron deficiency anemia in pregnant women by means of histochemical methods combined with computer microspectrophotometry.

Introduction: Decidua cells are important cells to the placenta, playing a significant role both in the physiology of pregnancy and during inflammation. The processes of oxidative modification of proteins (OMB) in inflammation are associated with increased levels of oxygen free radicals, which alter the properties of these macromolecules while oxidating amino groups of proteins. Anemic condition is accompanied by intensification of free radical processes in the blood and tissues, and iron deficiency additionally significantly modifies these processes.

Methods: 125 studied placentas, to compare the studied placental physiology of pregnancy and monitoring iron deficiency anemia without inflammation.

A histochemical reaction of bromophenol blue for "acidic" and "basic" proteins by Mikel Calvo was set in histological sections 5 μm thick.

Delta Optical Evolution 100 and Olympus SP-550UZ were used to obtain a digital copy of the image. Ratio R/B, which is the ratio between the amino and carboxyl groups in proteins, was determined by "ImageJ".

Unpaired Student's test calculated arithmetic mean and its error.

Results: When assessing visual histochemical preparations decidua cells are clearly stained, that is suitable for quantitative research, cell boundaries are defined through clear cell membrane coloring and contrasting color around decidua cells fibrinoid. Nuclei and nucleoli were visualized fairly well. "Basic" proteins prevailed in nucleoplasm, while "sour" in the nucleolus.

The decidua cells' cytoplasm specific color has been mostly granular in nature, and spectral characteristics and optical density of color varied greatly.

Factor R/B at physiological pregnancy ($n = 20$) was – 1.04 ± 0.008 and in iron deficiency anemia ($N = 21$) – 1.06 ± 0.009 $P > 0.05$. In acute chorioamnionitis ($n = 23$) – 1.08 ± 0.009, and combined with iron deficiency anemia ($N = 21$) – 1.09 ± 0.009 $P > 0.05$. Regarding chronic chorioamnionitis ($n = 20$) ratio – 1.24 ± 0.011, and combined with iron deficiency anemia ($N = 21$) – 1.64 ± 0.016 $P < 0.001$.

Conclusion: Conclusion. The intensity of OMB increases only in chronic form of chorioamnionitis in the decidua cells cytoplasm, and combined with iron deficiency anemia significant performance increase has been observed.

http://dx.doi.org/10.1016/j.pbj.2017.07.095

PS121

Comparison of Ras/Raf/MAPK signaling pathway in primary tumour and lymph node metastases – A report on an experimental study of two colorectal cancer cell lines (SW480 and SW620) and tissue samples

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Aim: To compare the presence of mutations in essential genes of CRC pathogenesis pathway between tissues derived from the primary tumour site and lymph node metastases.

Introduction: Colorectal cancer (CRC) remains the third most commonly diagnosed malignancy worldwide and a leading cause of cancer - related death. One of the pivotal pathways leading to CRC development is Ras/Raf/MAPK which is regulated by the receptor for the EGF. Mutations in these genes predict lack of response to EGFR-targeting monoclonal antibodies. However it is a common practice to assess only the primary tumour site, while mutations in metastasis may also affect the response to treatment.

Methods: The study was conducted on 10 patient-derived tissue samples and two ATCC human CRC cell lines obtained from the same individual: SW480 (primary tumour) and SW620 (lymph node metastasis). Cell lines were cultured according to the protocol. Genomic DNA and RNA were isolated, and PCR and RT-PCR were conducted. Primers for PCR included the following fragments: KRAS (exons 2,3,4), NRAS (exons 2,3,4), BRAF (exon 15); and for RT-PCR: KRAS, NRAS, BRAF and EGFR. Restriction enzymes were used. Proteins were extracted, purified and Western-Blot (RAS, RAF, MAPK) was performed.

Results: For SW480 we detected a mutation in exon 3 of NRAS gene, whereas SW620 presented a wild type. The level of Ras protein remained the same. Raf protein expression was abundant in the primary tumour site as compared to the lymph node metastasis, whereas MAPK protein presented the opposite level of expression.

Conclusion: The analysis of Ras-Raf-MAPK pathway may suggest that along with the tumour progression, the dominating signal is located at deeper levels of signaling pathway. Due to existing differences in key molecular points between the primary tumour and its metastases, in the era of targeted therapy, pre-treatment assessment of both sites has a potential to become a standard of care.[1,2]

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http://dx.doi.org/10.1016/j.pbj.2017.07.096

PS122

PI3K-Akt and Ras-Raf-MAPK signaling in colorectal cancer – Comparison of activity in primary tumor tissues and primary tumour – Derived human colorectal cancer cell lines