SUMMARY

The term "ovarian cancer" refers to neoplastic lesions that are derived from ovarian surface epithelial cells (OSE). This disease is one of the most common (commonest among cancers of the reproductive system) cause of women's deaths. In Poland, in 2015, ovarian cancer (OC) was the fifth in cancer incidence in women, while in terms of the number of deaths, it was fourth (after lung, breast and colon cancers). The most common types of ovarian cancer are: serous, mucous, endometrial and clear cell carcinoma. Other types of tumours (eg. squamous, undifferentiated, mixed) are diagnosed much less frequently.

Complement is not only important branch of anti-infective immunity – it also participates in tissue regeneration processes, removal of immune complexes, cells undergoing apoptosis and necrosis, can enhance cell proliferation and activate protooncogenes. It can be activated *via* classical, alternative and lectin pathways, which characteristic components (some collectins, ficolins, MASP proteases) are considered important factors of the innate immunity. The deficiency of mannose-binding lectin (MBL) is known as the most common immunity disorder in human. Ovarian cancer, due to the lack of specific symptoms at the early stage of the disease, is usually diagnosed at an advanced stage with unfavourable prognosis. For years, the primary marker of ovarian cancer has been the CA-125 glycoprotein, however, its level appears normal in up to 50% of Grade I cases (according to the World Organization of Gynecology and Obstetrics, *fr*. *Fédération internationale de gynécologie et d'obstétrique,* FIGO). Research has been conducted for a long time in order to find an ideal OC marker, quickly detectable in easily available material such as blood or urine. Despite significant achievements and the introduction of tests such as ROMA, Ova1 or Overa, it is still "... regrettable that we are still looking for effective methods for early diagnosis of cancer ..." [after: Ueland, FR, A Perspective on Ovarian Cancer Biomarkers: Past, Present and Yet-to-Come. Diagnostics (Basel), 2017. **7** (1)].

The aim of the study was to examine the significance of selected innate immunity factors associated with complement activation *via* the lectin pathway: mannose-binding lectin, ficolin-2, ficolin-3 and MASP-2 serine protease, in ovarian tumours.

Patients with diagnosed primary ovarian cancer (OC, n=128) and women qualified to the control group (C) [diagnosed with benign lesions (BT, n=123) and those who had not neoplastic changes (NO, n=74)] were recruited. The serum concentrations and/or activities of afore-mentioned factors were determined, and selected polymorphisms of the corresponding genes were investigated. The differences in median protein levels between groups were evaluated as well as the frequency of low and high concentrations in these groups, and the association of these values with genotypes. Moreover, the expression of the *MBL2, FCN2, FCN3 and MASP2* genes in selected organs of the female reproductive system was examined at the mRNA level and the relative amount of selected immune factors in tissue sections taken from unchanged and malignant ovaries was estimated. The expression of the analyzed proteins in ovarian and liver cancer cell lines was also examined.

It was shown that the median serum concentrations of ficollin-2 and ficolin-3 were significantly higher in the group of women diagnosed with primary ovarian cancer than in the reference groups, whereas there was no differences in the case of MBL and MASP-2, as well as the activity of MBL-MASP-2. However, it was observed that the frequency of genotypes associated with MBL deficiency (LXA/O or O/O) is significantly higher within the OC group compared with controls. This result suggests that deficiency of mannose-binding lectin may be a risk factor for the development of malignant changes in this organ. On the other hand, ROC analysis showed, that this primary deficiency promotes better prognosis for the patients. That may reflect partial weakening of the inflammatory response which is known to promote tumour progression. In women suffering from cancer who had such a disorder of immunity, the probability of a three-year survival was by 30% higher than in patients with genotypes other than LXA/O or O/O. Among the A/A homozygotes (genotype related to high *MBL2* gene expression and the synthesis of fully active MBL), the median concentration of this lectin (and the activity of its complexes with MASP-2) determined for the OC patients was significantly higher than for women included into the reference group. This may indicate that complement activation *via* the lectin pathway promotes carcinogenesis or, contrary, MBL concentration increases in response to the development of malignant tumour.

The selected polymorphisms of the following genes: *FCN2* (determined using in-house developed methods), *FCN3, MASP2* have no significant impact on the risk for developing ovarian cancer or prognosis for patients.

The intensity of immunostaining of ovarian histological sections with malignant lesions was assessed to be much higher than in the case of sections with benign tumours or unchanged

organs. This observation concerned all of the tested proteins. Both in the changed and unchanged ovaries, expression of the examined genes was found at the mRNA level. Expression of the *MBL2* and *MASP2* genes was higher, whereas *FCN2* and *FCN3* - lower in the samples obtained from malignant tumours than in samples from ovaries without neoplastic changes and/or with benign lesions. It was also observed that the level of *FCN3* gene expression at the mRNA level tested in ovarian sections could be considered as potentially useful in the OC diagnosis (in contrast to the level of *MBL2*, *MASP2* or *FCN2* gene expression). However, to fulfill requirements of usefulness, it would be necessary to have the possibility to take clinical material/perform test in a non-invasive way.

The presence of the tested proteins was demonstrated in ovarian and hepatic tumour cell lines, both by means of fluorescence microscopy (ficolin-2, -3, MBL) and/or flow cytometry (ficolin-2, -3, MASP-2). The obtained results suggest that continuation of the work is advisable and extension of its scope (other complement factors of the lectin pathway, such as ficolin-1, collectin-10 and -11 or other, enzymatic and non-enzymatic proteins of the MASP family) seems reasonable.