

***Characteristics of mammary gland cancer
associated macrophages in the context of the vitamin D influence on the tumor
microenvironment***

SUMMARY

The main objective of this study was to characterize macrophages associated with metastatic and non-metastatic breast cancers (TAMs) in the context of the influence of vitamin D₃ and its biologically active form – calcitriol – on their pro- or anti-cancer polarization status.

In vivo studies showed that both, the administration of food supplemented with vitamin D₃ and the treatment of mice with calcitriol, regardless of the food administered, increased the metastatic potential of 4T1 cells, which was not observed in the case of the 67NR and E0771 cell lines. It was also shown that in mice with an increased number of metastases to the lungs and bones, an increase in the pro-cancer phenotype of TAMs 4T1 - M2 (MHCII^{low}CD44^{high}) was observed. Simultaneously, TAMs 4T1 showed pro-inflammatory gene expression profile (increased expression of *Il23a* and decreased expression of *Il10*), which correlated with an increased level of the osteopontin in the tumor tissue. In addition, macrophages derived from the bone marrow of mice fed with diet deficient in vitamin D₃ and treated with calcitriol were more sensitive to inflammation-inducing factors secreted by calcitriol-stimulated 4T1 cells *ex vivo*. In addition, an increase in the level of CD44 (an osteopontin receptor) was observed in TAMs isolated from postoperative tissues of patients with locally advanced breast cancer. In the group of patients with normal levels of 25(OH)D₃ in plasma, a tendency to decrease the level of CD44 on the surface of TAMs was demonstrated. It was also shown that calcitriol modulated monocytes-derived macrophages by reducing the level of molecules characteristic of the M1 and M2 macrophage class, depending on the level of 25(OH)D₃ in plasma, the menopausal status of patients and the stage of the disease. In addition, in a group of premenopausal patients with normal plasma 25(OH)D₃ levels, an increased ability of these cells to polarize towards M1, while a decrease to polarize towards M2c class was observed.

In conclusion, the response of macrophages to the stimulation of particular factors directing their polarization depends on the vitamin D status of the organism from which they originate. However, the final polarization status of macrophages seems to depend mainly on the individual characteristics of cancer cells and their response to calcitriol, as cancer cells and the factors secreted by them predominate in the tumor microenvironment, affecting its composition and functioning.