https://doi.org/10.46389/rjd-2023-1298
www.rjdnmd.org

Rom J Diabetes Nutr Metab Dis
2023; volume 30, issue 1, pages 34-41

Original Article

Prognostic value of the level of expression of cell wall proteins and cytokines during inflammation in tumor tissue culture in patients with type 2 diabetes mellitus

Oleksii Volodimirovich Movchan [1], Irina Yuriivna Bagmut [2], Michael Ivanovich Sheremet [3] *, Lidia Anatolyivna Sukhanova [2], Igor Leonidovich Kolisnyk [2], Oleksandr Vasyliovych Bagmut [4], Olexksii Oleksandrovich Halmiz [5], Olena Valeriyivna Havrish [5], Andriy Oleksandrovich Lyashenko [1], Irina Viktorivna Dosenko [1], Oksana Mykolaivna Ivankova [1], Anton Dmitrovich Loboda [1], Oleksandr Volodimirovich Lazaruk [6]

[1] National Cancer Institute of the Ministry of Health of Ukraine, Kiyv, Ukraine

[2] Department of Infection Control, Kharkiv National Medical University of the Ministry of Health of Ukraine, Kharkiv, Ukraine

[3] Department of Surgery No.1, Bukovinian State Medical University, Chernivtsi, Ukraine

[4] Faculty of Computer Sciences, Karazin Kharkiv National University, Kharkiv, Ukraine

[5] Department of Neurology, Narcology, Psychiatry, Ophthalmology, Otolaryngology and Oncology of Luhansk State Medical University, Rivne, Ukraine

[6] Department of Pathology, Bukovinian State Medical University, Chernivtsi, Ukraine

* Correspondence to: Michael Ivanivich Sheremet, Department of Surgery No.1, Bukovinian State Medical University, Holovna str. 191, 58018, Chernivtsi, Ukraine. Phone: 0956064607; E-mail: mihayl71@gmail.com

Received: 7 January 2023 / Accepted: 21 February 2023

Abstract

The levels of chitinase-like proteins in circulating blood increase with inflammatory types of tumors.The aim of the study aimed to prognostic value of expression of chitinase-like proteins and interleukins levels in tumor tissue culture in patients with inflammatory breast cancer, diffuse stomach and diffuse esophagus cancer with amplification erBb2 (3+) with type 2 diabetes mellitus. We determined the expression of CD68, a marker of M2 subpopulation RS1 (stabilin-1), chitinase-like proteins YKL-39 i SI-CLP and interleukins levels on comparison of amplification erBb2 in patients with Inflammatory Breast Cancer (IBC), Diffuse Stomach (DSC) and Diffuse Esophagus Cancer (DEC). In all patients were performed conditions of plasma glucose levels after operation. Ist block: 21 patients with a confirmed mutation of the Her-2/neu gene and amplification erBb2 (3+) with type 2 diabetes mellitus: IBC 7, DSC 7 and DEC 7. IInd (block): 21 patients IBC 7, DSC 7 and DEC 7, without amplification erBb2 and normal range of plasma glucose level. As a model for tumor-associated macrophages: stimulation of human primary macrophages by cytokine M2-polarization of IL-4 and tumor cell supernatants (CSF2RA for breast cancer, and Colo206-M for stomach and esophagus cancer). With overexpression, erBb2 and serum markers increase for DSC and DEC by 36% (P<0.005). The immune microenvironment in the tissue of the tumors is manifested by higher levels of IL-1β and IL-8 (3 times), lower levels of IL-18 (2 times) and higher density of CD3 + (10 times). Thus, studies have shown that patients with type 2 diabetes mellitus operated on for inflammatory cancers are formed with deeper immune deficiency.

Keywords: chitinase-like proteins, interleukins, tumor tissue culture, inflammatory breast cancer, diffuse stomach and diffuse esophagus cancer, type 2 diabetes mellitus.

© 2023 The Authors. Romanian Journal of Diabetes, Nutrition and Metabolic Diseases published by Sanatatea Press Group on behalf of the Romanian Society of Diabetes Nutrition and Metabolic Diseases. This is an open access article under the terms of the Creative Commons Attribution License (CC-BY-NC-ND 3.0).

Introduction

Although patient selection was based on erBb2 overexpression, not all patients benefited from trastuzumab therapy. Therefore, the erBb2 gene dosage effect might provoke increased biological aggressiveness and altered trastuzumab sensitivity [1]. Absolute erBb2 copy numbers and ErBb2/centromere 17 ratios ("R"), as a rule, measured by FISH analysis in tumors receiving trastuzumab-based treatment for Her-2/neu overexpressing breast cancer, stomach cancer and esophagus cancer [2]. There are mainly inflammatory and diffuse forms of the disease. High-level erBb2 amplification is associated with a shorter time to first metastasis. In recent years, data on the involvement of chitinase-like proteins in the functioning of tumors-associated macrophages have been obtained [3]. Chitinase-like proteins are produced by several types of cells and combine the properties of cytokines and growth factors [4].

Numerous conceivable molecular mechanisms are proposed to clarify the causal relationship between T2DM and many types of cancer. Inflammation-related conditions may be of prime significance in clarifying the connection; thus, the survey encourages interconnected states. It builds up the multi-faceted, bidirectional, and double relationship between diabetes, its treatment, and cancer type-specific rate, prognosis, and treatment [5].

Extremely modest treatment results of patients with oncological pathology of any localization already under the IIIrd stage force specialists to look for new criteria for early diagnosis and marker integral detection indicators of the disease and its complications [6].

Diabetes is one of the foremost vital inveterate conditions worldwide, and cancer is the foremost predominant death disease worldwide. A few sorts of investigations have been conducted to discover the interface between diabetes and its potential for expanding the chance of cancer [7].

So, assessing the cellular and humoral immune link is undoubtedly a prognostic basis for the formation of carcinogenesis [8], especially infiltrative forms of malignant neoplasms [9]. To improve the survival of patients, it is necessary to introduce new methodological approaches to diagnosing not only localization [10], stage of development, morphological, genetic and molecular-biological characteristics of the tumor [11] but also the features of local homeostasis in the tumor cell [12], taking into account the change in tumor-associated immunocompetent cells in the process of the transformation of normal in cancer [13].

In the studied statistical and literary data selected for a comparative study of localizations, it is clear that the average percentage of patients with amplification erBb2 (3+) is as follows: breast cancer amplification of 15–20% [14], stomach cancer 10–15% [15] esophagus cancer 20–24% [16], characterized, as is known, a more aggressive flow of the disease and a high risk of relapses [17].

Each of the numerous phenotypic subtypes of tumor-associated macrophages is formed under the influence of cytokine-specific actions in a tumor microenvironment [18]. The results indicate the antitumor effect of macrophages for stomach cancer, where tumor-associated macrophages correlate with longer survival [19]. The high plasticity of macrophages to change their polarization under the influence of various microenvironment conditions opens the prospects of directed differentiation of macrophages in an antitumor M1 phenotype or blocking of M2 polarization [20]. The levels of chitinase-like proteins in circulating blood increase with inflammatory diseases and various types of tumors [21]. In this case, macrophages are a source of chitinase-like proteins, as well as targets for their regulatory effects [22].

The tumor stroma compartment is potentially a complete depot of the cytokines associated with the molecular components of the intercellular amorphous substance, the source of which is immune-inflammatory infiltrate cells [23]. It is in the stroma of this type of protein and the glycosaminoglycans of the intercellular matrix, to a greater extent, can bind cytokines and growth factors and represent them by cellular elements of stroma, thus providing optimal cell-stromal interaction and macrophage accumulation [24]. That is, interleukins are known to be a factor in the invasiveness of malignant tumors [25].

Recent studies have conclusively established an essential role of the tumor microenvironment in cancer progression and metastasis. The tumor microenvironment has been demonstrated to be enriched for proinflammatory cytokines, chemokines, and growth factors [26].

Chronic hyperglycemia, in collaboration with the other metabolic distortions in patients with diabetes mellitus, can cause harm to different organ frameworks, driving the advancement of debilitating and life-threatening well-being complications, most conspicuous of which are microvascular and macrovascular complications driving a 2-fold to the 4-fold expanded hazard of cardiovascular infections. In this audit, we offer an outline of the pathogenesis, determination,

© 2023 The Authors

clinical introduction, and standards of administration of diabetes [27].

The study aimed to determine the expression of CD68, M2 marker of subpopulation RS1 (stabilin-1), chitinase-like proteins YKL-39 i SI-CLP and interleukins levels on comparison of amplification erBb2 in tumor tissue and plasma glucose level, as an influencing factor in the development of the immune system deficiency in patients with inflammatory breast cancer, diffuse stomach and diffuse esophagus cancer, and investigate the prognostic value of those markers.

Material and methods

The study is conducted according to the ethical principles of the Helsinki Declaration, GCP (Good Clinical Practice), and the Law of Ukraine "On medications". All patients were informed about the research and signed the agreement.

In the period from 2017 to 2020, there were surveyed and treated 42 patients with inflammatory breast cancer, diffuse stomach and diffuse esophagus cancer. The first group (block): 21 patients with a confirmed mutation of the Her-2/neu gene and amplification erBb2 (3+) with type 2 diabetes mellitus (patients blood glucose range over 10.0 mmol\l): breast cancer – 7; stomach – 7; esophagus – 7. The second group (block): 21 patients with inflammatory breast cancer –7, diffuse stomach – 7 and diffuse esophagus cancer – 7, without amplifying erBb2.

Tissue arrays for marker macrophage population CD68, M2 subpopulation marker RS1 (Stabilin-1), chitinase-like proteins YKL-39 and SI-CLP immunohistochemical analysis were available from 42 patients with inflammatory breast cancer, diffuse stomach and diffuse esophagus cancer – for each localization (median age 59 years, range 26–80). According to the classification of TNM tumors corresponding to the III-IV stage, they histologically confirmed adenocarcinoma G III-G IV.

For example, we studied the operational material of patients with stomach cancer. The average score of expression of markers studied in groups of patients is presented in Table 1.

Identification of erBb2 amplification is performed using fluorescence in situ hybridization (FISH) and immunohistochemistry (IHC) for HER-2 overexpression. Further characterization of DNA amplification can be performed using digital droplet PCR (ddPCR) and low-coverage whole genome sequencing (lcWGS). DdPCR is a robust and precise method for enumerating a specific DNA segment's copy number (CN). LcWGS identifies DNA amplifications and deletions throughout the genome as well as amplicon structure (AS). Combining these methods, we were able to detail an amplicon CN and AS.

Subsequently, we studied tumor-associated macrophages and the state of chitinase-like proteins of tumor stroma. As a model for tumor-associated macrophages, a system of stimulation of human primary macrophages was used by cytokine M2-polarization of IL-4 and tumor cell supernatants according to breast cancer, stomach cancer, and esophagus cancer. The expression of CSF2RA for breast cancer and Colo206-M for stomach and esophagus cancer and tumor biopsy was evaluated by immunohistochemical method.

Immunohistochemistry

Tumor biopsy samples (8 mm^3), obtained using core biopsy, 11 were washed with culture medium Dulbecco modified Eagle medium (DMEM)–F12 3 times to wash off the remaining blood cells on their surface and then were placed into a glass vial with 1 mL of the DMEM–F12 growth medium and incubated for 72 hours to accumulate in the supernatant sufficient (for an accurate assessment of each cytokine) concentration of all the cytokines studied by us. Before collection of the supernatant, the tumor biopsy samples were retrieved

Table 1: Average expression score of CD68, Stabilin-1, YKL-39, SI-CLP in tumor tissue culture with diffuse stomach cancer in groups with amplification erBb2 with type 2 Diabetes Mellitus and its absence.

Investigated indicators	Average expression score M±SD (N) in a group with amplification erBb2 (3+) with type 2 Diabetes Mellitus (7 patients)	Average expression score M±SD (N) in a group without amplification erBb2 (7 patients)
CD68	2.96±0.81 (n=33)	2.26±0.82 (n=38)
YKL-39	2.35±0.99 (n=17)	2.24±0.72 (n=37)
SI-CLP	2.63±1.24 (n=30)	2.23±0.77 (n=35)
Stabilin-1	2.94±1.01 (n=32)	2.0±0.79 (n=36)

Note: Reliable differences from the indicator: * – healthy tissue; ^ – tissue of the peritumoral zone ($p<0.05$).

https://doi.org/10.46389/rjd-2023-1298
© 2023 The Authors

from the vial and placed in 10% neutral formalin. After culturing the biopsy samples in the supernatant, there was a small number (no more than 50–100 cells for the whole supernatant) of cellular elements (single tumor, lymphoid, and monocytic cells) that were removed from the supernatant by precipitating with centrifugation at 900 × g for 15 minutes. By enzyme-linked immunosorbent assays, the concentrations of IL-2, IL-6, IL-8, IL-10, IL-17, IL-18, IL-1β, tumor necrosis factor α (TNF-α), IFN-γ, CSF2RA and Colo206-M for breast cancer of were determined in culture supernatants. The calculations were performed based on the manufacturer's recommendation. The results were shown as pg/ml.

Statistical analysis

Histograms were created in Microsoft Excel, Statistica version 7, and IBM SPSS Statistics version 22.0. Determination of mean values, medians, 25th to 75th percentiles, and Spearman rank correlation coefficient (r) was performed through the software package Statistica version 7. The cluster analysis was performed using Statistica version 7. Receiver operating characteristic (ROC) curve analysis was performed utilizing the software package IBM SPSS Statistics version 22.0.

Results

As an example, we took patients with stomach cancer with amplification ErBb2 that had the recurrent disease for 1 year since the beginning of special treatment (surgery + chemotherapy) and without amplification erBb2, which did not detect recurrence schedules for 1 year after a special treatment. It should be noted that when studied, the average expression of CD 68, Stabilin-1, YKL-39, and SI-CLP in tumor tissue inflammatory breast cancer and diffuse esophagus cancer results were almost identical (p=0.005).

Multivariate Cox analysis, including amplification erBb2, plasma YKL-39, serum CD 68, SI-CLP, Stabilin-1, TNM stage, age and radicality after primary surgery as variables, showed that elevated plasma YKL-39, serum CD 68, SI-CLP, Stabilin-1 was associated with shorter survival (HR=2.13, 95% CI: 1.40–3.25, p=0.0004).

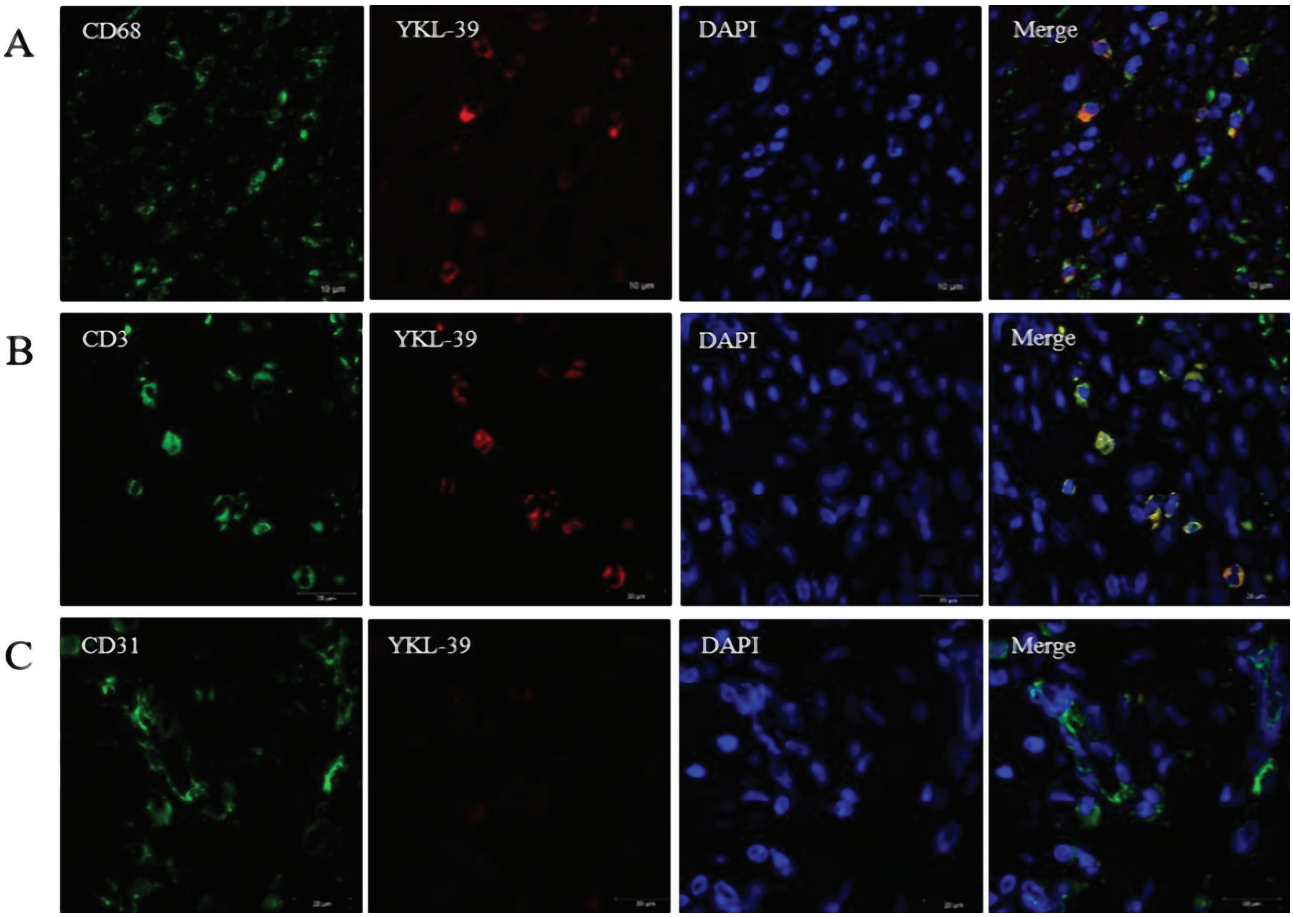

Figure 1: Immunofluorescence analysis of YKL-39 expression in stomach cancer samples in various tumor microenvironment cells: A – CD68-positive macrophages; B – CD3 + T-lymphocytes; C – blood vessels.

© 2023 The Authors

The interconnection of the expression level of the tested proteins YKL-39, Si-CLP, CD68, and Stabilin-1 with the presence or absence of recurrence in the stomach cancer was not detected. In patients with stomach cancer without of amplification, erBb2 expression YKL-39 was statistically significant in a group of patients with amplification erBb2 with type 2 diabetes mellitus (1.68±0.71, p=0.03) compared to a group without amplification erBb2 and without diabetes (2.73±0.87, p=0.044).

In a group of patients without amplification erBb2, with all localization studied in our investigation, expression of YKL-39 remained statistically significantly lower in the presence of recurrences (1.79±0.81, n=13 *vs.* 2.47±0.55, n=22, p=0.024).

In patients with inflammatory breast cancer, diffuse stomach and diffuse esophagus cancer, there was a tendency to increase the expression of YKL-39 with the presence of amplification erBb2 with type 2 diabetes mellitus in inflammatory tumor infiltration located in the stroma. A higher expression of stabilin-1 (2.1±0.70, n=21) was detected compared to tumors of patients who did not have amplification erBb2 (1.46±0.67, n=10, p=0.015).

For YKL-39 on the material from patients with stomach cancer, an analysis of the localization of the expression of tumor tissue was carried out, which was found that YKL-39 is expressed in CD68-positive tumor-associated macrophages and CD3 + T-lymphocytes (Figure 1).

The patient's inflammatory breast cancer with overexpression erBb2 and serum marker of macrophage population CD68, a marker of M2 subpopulation RS1 (stabilin-1), chitinase-like proteins YKL-39 i SI-CLP detected in 30% (p<0.0001). These patients were sicker (p<0.01) and more often had parenchymal involvement (p<0.0005). With overexpression erBb2 and serum of M1 marker of macrophage population CD68, a marker of M2 subpopulation RS1 (stabilin-1), chitinase-like proteins YKL-39 i SI-CLP detected in the stomach and esophagus cancer in 36% (p<0.005).

Consequently, several differences in the level of factors of local immunity are established in patients with inflammatory breast cancer and diffuse stomach and esophagus cancer. The higher levels of proinflammatory cytokines are shown, preferably IL-1 and IL-8, in tumor tissue compared without amplification erBb2.

The group of immune-score research consisted of 21 patients with inflammatory breast cancer, diffuse stomach and esophagus cancer with amplification erBb2 (3+) with type 2 Diabetes Mellitus. The average concentration of CD3 cells in tumor stroma (343±81) (interval 32–455, mode 415, median 205) of T-lymphocytes per 1 mm^2; CD3 cells in the central parts of the tumor per 1 mm^2 (215±57); CD8 T-lymphocytes in tumor stroma (144±114) cells per 1 mm^2; CD8 cells in central sections – (122±104) T-lymphocytes per 1 mm^2; CD68 M1 tumor macrophages – (263±98) cells in tumor stroma per 1 mm^2; CD68 macrophages in the central parts of the tumor (293±125) cells per 1 mm^2; CD163 M2 macrophages in a stroma (144±86) cells per 1 mm^2; CD163 M2 of macrophages in the central parts of the tumor – (142±127) units per 1 mm^2, *in vitro*. The ratio of CD3/CD8 T-lymphocytes, which reflects the intensity of the immune response of the body to the tumor was 2.21 (Figure 2).

In total, in the process of experimental study, specific cells were found – 25495, of which of macrophages – 13829, and T-lymphocytes – 11666. CD3 prevailed in tumor stroma (3333 from 3919 of all detected), CD8 T-killers also prevailed in tumor stroma (1967 out of 2447 of all detected), CD68 M1 macrophages, on the contrary, prevailed in the tumor "nest" (4314 out of 4760 of all detected) and CD163 M2 macrophages also prevailed in the central parts of the tumor (2347 of 2408 of all detected during the process). Only the saturation of macrophages in the tumor "nest" correlated with a favorable forecast. In the red group of patients with immune-score equalized 1 or 2 (immune tissue response is present), in blue – 0 (immune tissue answer is absent), p<0.05 (Figure 2).

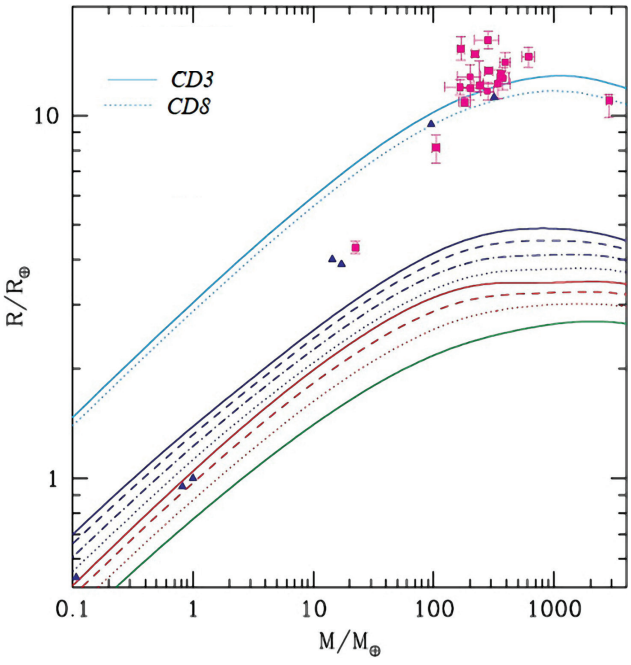

Figure 2: The ratio of CD3/CD8 T-lymphocytes reflects the intensity of the immune response of the organism to the tumor *in vitro*.

https://doi.org/10.46389/rjd-2023-1298
© 2023 The Authors

Table 2: The specific weight of some proinflammatory cytokines in tumor tissue *in vitro* patients with inflammatory breast cancer, diffuse stomach and esophagus cancer with amplification erBb2 (3+) with type 2 Diabetes Mellitus.

Investigated indicators, N	Cytokines levels (pg/ml/g protein)				
	IL-1ß	IL-6	IL-8	IL-18	TNF-α
Breast cancer, 7	48.8±8.5*	13.5±2.2	50.0±6.5*	164.8±32.1**	2.4±0.4
Stomach, 7	67.9±32.2	26.8±17.1	48.8±15.0	311.2±32.3	2.1±0.4
Esophagus, 7	15.7±6.3	18.5±10.8	17.2±7.6	257.1±66.5	1.7±0.2

Note: * – statistically significant differences from the indicator of healthy tissue; ** – statistically significant differences from the tissue of the peritumoral zone (p<0.05).

In the red group of patients, the CD3/CD8 ratio was more than 1.6 (immune tissue response is more pronounced), and in the blue group – less than 1.6 (immune tissue response is weak or absent), p<0.05.

At the end of the follow-up, a total of 42 patients had died, 28 (median follow-up time: 14 months, range: 3–31), and 14 patients were still alive (median follow-up time: 18 months, range: 8–36). Multiple Cox regression analysis revealed three independent predictors of the patient's overall survival (HR=0.19, 95%, CI 0.14–0.49, p=0.0005).

Discussion

The presence of chitinase-like proteins and increasing the index of their proliferation during the presence of erBb2 amplification and plasma glucose level above 10.0 mmol/l as a confirmation marker indicates the aggressiveness of the cancer process. However, in the future, and we already conduct such work, the presence and saturation of chitinase-like proteins of the tumor microenvironment may be a factor in higher sensitivity to special treatment with selected localizations. We showed that the "cytokine profile" of a tumor (the ability of tumor cells and its microenvironment to produce different cytokines) is very individual, especially when its present amplification erBb2 plasma glucose level is above 10.0 mmol/l. These results can be useful when choosing an individualized treatment with selected tumor localizations (both system and targeted). It has been shown that the features of the cytokine profile of mammary adenocarcinoma are important for the formation and realization of the metastatic potential of mammary adenocarcinoma and diffuse stomach and diffuse esophagus cancer. According to this profile, it is possible to predict the development of complications at the stages of the complex treatment of cancer and the possible development of relapses. In addition, the expression of YKL-39 has negatively correlated with the number of lymph nodes with metastases (R=-0.89). When studied in interleukins levels in the culture of tumor tissue in patients with inflammatory breast cancer, diffuse stomach and diffuse esophagus cancer with amplification erBb2, significant differences are found, depending on the localization of the oncology process,

Table 3: Immunohistochemical indicators of interleukins in tumor tissue culture in patients inflammatory breast cancer, diffuse stomach and esophagus cancer.

Interleukins	I group [with amplification erBb2 (3+)]			II group (without amplification erBb2)		
	Breast cancer, 7	Stomach, 7	Esophagus, 7	Breast cancer, 7	Stomach, 7	Esophagus, 7
IL-1	43.3±4.1^•	49.9±5.16^	58.5±4.4*•^	270.3±25.5	80.29±9.95*•	100.1±13.1*
IL-2	35.7±3.1^	32.6±3.7^	60.3±6.6*•^	18.1±3.31	7.65±1.42*	5.55±0.74*•
IL-6	21.9±4.7^	25.5±5.1^	39.11±4.4^*	190.9±23.1	16.8±1.22*•	55.21±4.6*•
IL-8	181.1±16.7^	166.2±14.^	169.4±11.3^	520.1±19.0	242.5±20.4*	130.2±12.7*•
IL-10	26.6±2.5^	15.5±1.8*^	17.4±1.9*^	8.56±1.94	6.22±1.55*	8.24±1.75
TNF-α	7.08±1.13^	5.9±1.05^	5.33±0.91*^	17.32±1.12	11.73±1.51*	12.07±0.65*

Note: The reliability of the differences: * – regarding conditionally healthy tissue: * – according to Student's criterion, p<0,05; • – by Z-criterion, p<0.05; ^ – relative to the relevant sections of the comparison group.

© 2023 The Authors

indicating a different index of tumor invasiveness at inflammatory breast cancer, diffuse stomach and diffuse esophagus cancer, that is, local distribution or infiltrative growth (Table 2). Most likely, more pronounced activity in the hollow organs depends on the constant mechanical load, including enzymatic in the process of digestion. That is, for all localizations selected for the study, the level of proinflammatory agents, with erBb2 amplification, is lower, which indicates an insufficient immune response (launching of protective forces), so fast cancer progression (Table 3).

Conclusions

Overexpression erBb2 and serum of macrophage population marker CD68, M2 subpopulation RS1 (stabilin-1), chitinase-like proteins YKL-39 i SI-CLP independently identified subgroups of patients with inflammatory breast cancer, diffuse stomach and diffuse esophagus cancer with a poor prognosis. Invasive activity of tumor cells with amplification of erBb2 for selected localization studies: breast cancer is lower than in stomach cancer and esophagus, proved by the levels of interleukins in the tissue culture.

Changes in the immune microenvironment in the tissue of the stomach adenocarcinoma (*in vitro*) with amplification of erBb2 are manifested by higher levels of proinflammatory IL-1β and IL-8 (3 times) and lower levels of IL-18 (2 times), higher density of CD3 + (10 times). CD19 + (4 times) lymphocytes and lower density CD3 + CD8 + (1.5 times) and CD16/56 + (2 times) of lymphocytes compared without amplification of erBb2 ($p<0.05$).

In patients with the presence of amplification erBb2 in inflammatory tumor infiltrate located in the stroma, the higher expression of chitinase-like protein YKL-39 and M2 marker of stabilin-1 macrophages is detected compared to patient tumors without amplification erBb2. Expression of YKL-39 in patients with stomach cancer and amplification erBb2 with the presence of lymph node lesions and metastases are significantly lower compared to a group of patients without metastases and negatively correlate with the number of lymph nodes with metastases (R=-0.89).

Despite the fact that patient selection based on erBb2 overexpression, not all patients benefit from trastuzumab therapy (target therapy), especially with type 2 diabetes mellitus. Patients with amplification erBb2 (3+) are as follows: breast cancer amplification of 15–20%, stomach cancer 10–15%, esophagus cancer of 20–24%, characterized, as is known, a more aggressive flow of the disease and a high risk of relapses. Patients with amplification erBb2 (3+) in combination with inflammation have a worse prognosis for cancer disease treatment.

This study demonstrates the important role of both quantities and phenotype tumor-associated macrophages in the progression of the tumor depending on the availability of amplification erBb2 and the presence or absence of type 2 diabetes mellitus. Therefore, we now understand the influence of specific proinflammatory factors and can add specific points against them in treating inflammatory and diffuse forms of cancer to reduce the level of the inflammatory response and, accordingly, the best chances of survival for patients.

Conflict of interest

The authors declare no conflict of interest.

Ethics approval

The approval for this study was obtained from the Bioethics Committee of Ukraine's National Cancer Institute (Minutes No. 168, July 24, 2021).

References

1. American Society of Clinical Oncology. Breast Cancer Guide. July 2019. Available at: www.cancer.net/cancer-types/breast-cancer/introduction
2. Boldrin E, Mazza M, Piano MA, Alfieri R, Montagner IM, Magni G, Scaini MC, Vassallo L, Rosato A, Pilati P, Scapinello A, Curtarello M. Putative Clinical Potential of ERBB2 Amplification Assessment by ddPCR in FFPE-DNA and cfDNA of Gastroesophageal Adenocarcinoma Patients. Cancers (Basel). 2022; 14(9):21802192. doi: 10.3390/cancers14092180. PMID: 35565309; PMCID: PMC9102116. https://pubmed.ncbi.nlm.nih.gov/35565309/
3. Kadomoto S, Izumi K, Mizokami A. Macrophage Polarity and Disease Control. Int J Mol Sci. 2021; 23(1):144-154. Published 2021 Dec 23. doi:10.3390/ijms23010144. https://pubmed.ncbi.nlm.nih.gov/35008577/
4. Tsuruta S., Kohashi K., Yamada Y., Fujiwara M. Solid-type poorly differentiated adenocarcinoma of the stomach: Deficiency of mismatch repair and SWI/SNF complex.Cancer Sci. 2020. Mar 11; 1(3):1008-1019. doi: 10.1111/cas.14301. Epub 2020 Jan 28. PMID: 31922331; PMCID: PMC7060473. https://pubmed.ncbi.nlm.nih.gov/31922331/
5. Durrani, I.A., Bhatti, A. & John, P. The prognostic outcome of "type 2 diabetes mellitus and breast cancer" association pivots on hypoxia-hyperglycemia axis. Cancer Cell Int. 2021; 21:351-357. https://doi.org/10.1186/s12935-021-02040-5

 https://doi.org/10.46389/rjd-2023-1298 © 2023 The Authors

6. Bhat, A.A., Nisar, S., Maacha, S. *et al*. Cytokine-chemokine network-driven metastasis in esophagus cancer; a promising avenue for targeted therapy. Mol Cancer 20, 2 (2021). https://doi.org/10.1186/s12943-020-01294-3
7. Eketunde AO. Diabetes as a Risk Factor for Breast Cancer. Cureus. 2020;12(5):e8010. Published 2020 May https://doi:10.7759/cureus.80105
8. Blank S, Nienhüser H, Dreikhausen L, Sisic L, Heger U, Ott K, Schmidt T. Inflammatory cytokines are associated with response and prognosis in patients with esophagus cancer. Oncotarget. 2017 Jul 18; 8(29):47518-47532. doi: 10.18632/oncotarget.17671. PMID: 28537901; PMCID: PMC5564583. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5564583/
9. Hoenerhoff M, Boyle M, Minocherhomji S, Pandiri S. Chapter 8 - Carcinogenesis: Mechanisms and Evaluation, Haschek and Rousseaux's Handbook of Toxicologic Pathology (Fourth Edition), Academic Press, 2022;1:205-254, ISBN 9780128210444, https://doi.org/10.1016/B978-0-12-821044-4.00013-3. https://www.sciencedirect.com/science/article/pii/B9780128210444000133
10. Méndez-García L, Nava-Castro K, Ochoa-Mercado T, Palacios-Arreola M, Ruiz-Manzano R, and al. Metastasis: Are Cytokines Important Players During Its Development and Progression?-Montor.Journal of Interferon & Cytokine Research Volume: 39 Issue 1: January 10, 2019.39-5. doi.org/10.1089/jir.2018.0024. https://europepmc.org/article/med/30321090
11. Ning-Bo Hao, Mu-Han Lü, Ya-Han Fan, Ya-Ling Cao, Zhi-Ren Zhang, Shi-Ming Yang, "Macrophages in Tumor Microenvironments and the Progression of Tumors", Journal of Immunology Research, vol. 2012, ArticleID 948098, 11 pages, 2012. doi.org/10.1155/2012/948.098. https://cyberleninka.org/article/n/472526
12. Wang X, Zheng Z, Sun Y *et al*. Significance of HER2 protein expression and HER2 gene amplification in colorectal adenocarcinomas. World J Gastrointest Oncol. 2019; 11(4):335-347 doi: 10.4251/wjgo.v11.i4.335. PMID: 31040898; PMCID: PMC6475672. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6475672/
13. Yao M, Brummer G, Acevedo D, Cheng N. Cytokine Regulation of Metastasis and Tumorigenicity. Adv Cancer Res. 2016;265-367. doi: 10.1016/bs.acr.2016.05.005. Epub 2016 Aug 5. PMID: 27613135. https://pubmed.ncbi.nlm.nih.gov/27613135/
14. American Cancer Society. Breast Cancer Facts & Figures 2019-2020. Available at: www.cancer.org/content/dam/cancer-org/research/cancer-facts-and-statistics/breast-cancer-facts-and-figures/breast-cancer-facts-and-figures-2019-2020.pdf
15. Azarhoosh R, Ebneghasem R, Besharat S. HER-2/neu gene amplification in gastric adenocarcinoma and its relationship with clinical and pathological findings. J Gastrointest Oncol. 2017; 8(6):1046-1050. doi: 10.21037/jgo.2017.10.11. PMID: 29299365; PMCID: PMC5750194. https://pubmed.ncbi.nlm.nih.gov/29299365/
16. Maoz M, Devir M, Inbar M, Inbar-Daniel Z, Sherill-Rofe D *et al*. Clinical Implications of Sub-grouping HER2 Positive Tumors by Amplicon Structure and Co-amplified Genes. Sci Rep. 2019; 9(1):187-195. doi: 10.1038/s41598-019-55455-6. Erratum in: Sci Rep. 2020;10(1):3941. PMID: 31827209; PMCID: PMC6906288. https://pubmed.ncbi.nlm.nih.gov/31827209/
17. Franzè E, Stolfi C, Troncone E, Scarozza P, Monteleone G. Role of Interleukin-34 in Cancer. Cancers (Basel). 2020; 12(1):252. Published 2020 Jan 20. doi: 10.3390/cancers12010252. PMID: 31968663; PMCID: PMC7017118. https://pubmed.ncbi.nlm.nih.gov/31968663/
18. Hu H, Ai, Lu M, Song Z, Li H. Characterization of intratumoral and circulating IL-10-producing B cells in gastric cancer. Exp Cell Res. 2019; 384:1116-1152. doi: 10.1016/j.yexcr.2019.111652. Epub 2019 Sep 28. https://pubmed.ncbi.nlm.nih.gov/31574287/
19. Bagmut I, Movchan O, Sheremet M, Smolanka I, Lyashenko A, Dosenko I, Loboda A, Characteristics of certain genetical and biological properties of carcinogenesis in the development of inflammatory breast cancer with type 2 diabetes mellitus and tumor relapse. Rom J Diabetes Nutr Metab Dis 2022;29 (2): 245-252. https://doi.org/10.46389/rjd-2022-1082
20. Vrána D, Matzenauer M, Neoral Č, Aujeský R, Vrba R *et al*. From Tumor Immunology to Immunotherapy in Gastric and esophagus Cancer. Int. J. Mol. Sci. 2019;20: 13-32. doi: 10.3390/ijms20010013. PMID: 30577521; PMCID: PMC6337592. https://pubmed.ncbi.nlm.nih.gov/30577521/
21. Zhao T, Su Z, Li Y, Zhang X, You Q. Chitinase-3 like-protein-1 function and its role in diseases. Signal Transduct Target Ther. 2020; 5(1):201. Published 2020 Sep 14. doi:10.1038/s41392-020-00303-7. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7490424/
22. Movchan O, Bagmut I. Impacts of Inflammatory Microenvironment on the Development of Cancer Relapse after Combined Treatment. Ukraïns'kij žurnal medicini, biologiï ta sportu. 2021; 6: 204-213. doi: 10.26693/jmbs06.05.204. https://pdfs.semanticscholar.org/3d72/100d33705abcd740db7755f14817f55e5562.pdf
23. Lopez-Soto A, Gonzalez S, Smyth M, Galluzzi L. Control of metastasis by NK cells. Cancer Cell. 2017;32:135–154. doi: 10.1016/j.ccell.2017.06.009. PMID: 28810142. https://pubmed.ncbi.nlm.nih.gov/28810142/
24. Qu X, Tang Y, Hua S. Immunological Approaches Towards Cancer and Inflammation: A Cross Talk. Front Immunol. 2018;9:563-572. doi: 10.3389/fimmu.2018.00563. PMID: 29662489; PMCID: PMC5890100. https://pubmed.ncbi.nlm.nih.gov/29662489/
25. Li Z, *et al*. Clinical significance of serum chemokines in esophagus Cancer. Med Sci Monit. 2019;25:5850–5855. doi: 10.12659/MSM.916846. PMID: 31385574; PMCID: PMC6693365. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6693365/
26. Morris R, Mortimer T, O'Neill K. Cytokines: Can Cancer Get the Message? Cancers (Basel). 2022;14(9):2178-2184. doi: 10.3390/cancers14092178. PMID: 35565306; PMCID: PMC9103018. https://pubmed.ncbi.nlm.nih.gov/35565306/#:~:text=Increasing%20evidence%20shows%20that%20cytokines,maintenance%2C%20invasion%2C%20and%20metastasis.
27. Goyal R, Jialal I. Diabetes Mellitus Type 2. [Updated 2021 Sep 28]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022 Jan-. Available from: https://www.ncbi.nlm.nih.gov/books/NBK513253

© 2023 The Authors