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VALUE OF CONDITIONALLY-PATHOGENIC MICROFLORA INDEX AS PREDICTING FACTOR OF BACTERIAL DYSBIOSIS' DEVELOPMENT

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Abstract

An article shows the most informative indicators that objectively reflect the condition of the pathological process of dysbiosis, and a system for predicting of the risk of occurrence and severity of dysbiosis according to these indicators is developed. Results of the study substantiated the assumptions about the pathogenetic role of disregulation of hormonal and immune systems in the occurrence of dysbiosis and bacterial vaginosis. Synshronously, the stage of the development of the immune system reaction is discovered in the during the flow of vaginal dysbiosis - from the control stay while normocoenosis to the immunoresistance in grade 1 dysbiosis and expressed combined immunodeficiency in the presence of specific humoral response to bacterial antigens in 2nd grade dysbiosis.

Key words: dysbiosis; conditionally pathogenic microflora; diagnostics; colonial resistance.

Dysbiosis is one of the most common types of infectious pathology of genitals in reproductive age women mainly. Its part among all vulva and vaginal infections of the female lower genital tract is from 12% to 80% [1]. The incidence of dysbiosis has doubled in the last decade and is, according to various authors, from 26% to 40-45% [2-4].

Dysbiosis is the most common cause of unusual vaginal discharge in women of childbearing age and occurs in 35% of women turning to dermatological and venereological clinics, in 15-20% of pregnant women, and in 5-15% of women seen by gynecologists [19-21]. In pregnant women from risk groups, dysbiosis is diagnosed in 25.0-40.3% of patients; nonspecific vaginitis – in 23.6-29.0% of patients; vaginal candidiasis –in 31.6-45.0% of patients [18].

To this day, a leading role of obligate anaerobesin the occurrence of dysbiosiswas convincingly proven; in this connection, dysbiosis is considered a polymicrobial vaginal syndrome and is characterized not only by vaginal discharge, but also by lesions of the cervix, body of the uterus, its appendages, as well as is the cause of pathologies of pregnancy and childbirth [5, 16].

Etiological structure of infectious agents has changed significantlyin the last decade due to the constant evolution of bacteria and involvementof conditionally pathogens in pathological processes [4]. Due to their dual nature, conditionally pathogens are present in the normal microflora of healthy people as commensals, and are also registered as etiopathogens in various local and generalized processes [13, 17]. Clinicians often have difficulties in estimation the examination results and determining the appropriateness of the prescribed treatment and choice of drugs [8-9].

Chronic inflammatory processes of the internal genitals in women should be considered as a general multisystem disease, accompanied by involvement in the pathological process of all parts of the neuroendocrine system, central and autonomic nervous system, cardiovascular, urinary, immune systems, hemostasis, and metabolism. This leads to impairment of the specific functions of the female organism (menstrual, sexual, reproductive), emergence of pelvic pain syndrome and generalization of the process [14-15].

Thus, determining the inflience of disorders of neuro-hormonal regulation, immune status and local factors of colonization resistance on the biocenosis of the vagina, will increase the efficiency of diagnostics and prediction of bacterial vaginosis.

Materials and methods

The analysis was performed for the results of previous examinations of 298 patients, 53 of whom were diagnosed with normocenosis, and 245 –with dysbiosis. 58 indicators were chosen as factor features (Table 1). [6, 7, 10-12].

At the first stage of the analysis, ICPM (index of conditionally pathogenic microflora) (variable Z) was considered as the resulting feature, at this, in the case of normocenosis

variable Z acquired the value Z = 0, in the case of grade I and II dysbiosis variable Z acquired the value Z = 1.

Table 1

Input signs of the primary analysis of indicators of colonization resistance of vagina, immune system and of hormonal regulation system

X1	Age	X20	IL10	X39	CD22
X2	MC day	X21	TNFα	X40	LPA
VS indicators:		X22	TGF-1β	X41	LPA in.
X3	IgM	X23	pH	X42	CIR
X4	IgA	Blood indicators:		X33	C3
X5	IgG	X24	FSH	X44	C4
X6	IgG_2	X25	LT	X45	γ-INF
X7	sIgA	X26	E_2	X46	IL1β
X8	Lysozyme	X27	PG	X47	IL2
X9	LPA	X28	TS	X48	IL4
X10	LPA in.	X29	CR	X49	IL6
X11	IC	X30	PRL	X50	IL8
X12	C3	X31	free T_3	X51	IL10
X13	C4,	X32	free T_4	X52	TNFα
X14	γ-INF	X33	LC	X53	TGF-1β
X15	IL1β	X34	CD16	X54	IgM
X16	IL2	X35	CD3	X55	IgA
X17	IL4	X36	CD4	X56	IgG
X18	IL6	X37	CD8	X57	IgG_2
X19	IL8	X38	IRI	X58	sIgA

Notes: MC – menstrual cycle; VS– vaginal secretion; LPA in.–Leucocyte Phagocytic Activity index; T_3 free– free T_3; T_4 free– free T_4; LC– lymphocytes; IRI – immune reactivity index; PRL – prolactin; CR – cortisol; TS– testosterone; FSH – follicle-stimulating hormone

To check the quality of the prediction model, all observations (using a random number generator) were divided into three sets: training (used to calculate the model parameters, 248 cases), control (used to control model retraining, 20 cases), confirmatory (used to check adequacy of the models when predicting on the new data, 30 cases).

Results and discussion. A linear neural network model was constructed and trained on a complete set of 58 factor features. Sensitivity of the model for predicting the risk of

dysbiosis according to ICPM based on a complete set of factor features was 100% (95% Confidence Interval 99.1% -100%), specificity – 100% (95% CI 95.6% -100%) on the training set, and 95.8% (95% CI 83.6% -100%) and 100% (95% CI 71.7% -100%) respectively on the confirmatory set.

Sensitivity and specificity on the training and confirmatory sets did not differ with any statistical significance ($p = 0.20$ and $p>0.99$, respectively, when compared by the χ^2criterion), which indicated the adequacy of the model being constructed.

To identify the factors most associated with risk the development of dysbiosis according to ICPM, significant traits were selected using a genetic selection algorithm. As a result, four factor traits were selected: the content of IL10 (X20) in the vaginal secretion, as well as the content of IL2 (X47), IL4 (X48) and IL6 (X49) in the blood.

A linear neural network model was constructed and trained on a selected set of four factor features. Sensitivity of the linear neural network model for predicting the risk of dysbiosis according to ICPM based on four factor features was 96.1% (95% VI 93.0% - 98.3%), specificity – 95.3% (95% CI 86, 8% -99.6%)on the training set, and 87.5% (95% CI 70.6% -97.8%) and 100% (95% CI 71.7% -100%) respectively on the confirmatory set.

Sensitivity and specificity on the training and confirmatory sets did not differ with any statistical significance ($p = 0.17$ and $p = 0.58$, respectively, when compared by the χ^2criterion), which indicated the adequacy of the model being constructed.

To estimate the significance of the selected factor features, the method of construction of operational characteristics curves (ROC-curves – Receiver Operating Characteristic Curve) of models was used (Fig. 1).

When performing the analysis, the area under the ROC-curve of the linear neural network model, constructedbased on all 58 factor features AUC1 = 1.00 (95% CI 0.99-1.00), differed with statistical significance ($p <0.001$) from 0.5.

The area under the ROC-curve of the linear neural network modelbased on four selected factor traits AUC2 = 0.99 (95% CI 0.97-1.00), differed with statistical significance ($p <0.001$) from 0.5. The analysis indicated a high significance of the selected factor traits (content of IL10 in vaginal secretion and content of IL2, IL4 and IL6 in blood) for predicting the risk of development of dysbiosis according to ICPM.

To identify the strength and direction of influence of the four selected factor features, a logistic regression model was constructed, and it proved to be adequate ($\chi^2 = 234.9$ at $p <0.001$). Results of analysis of the coefficients are given in table 2.

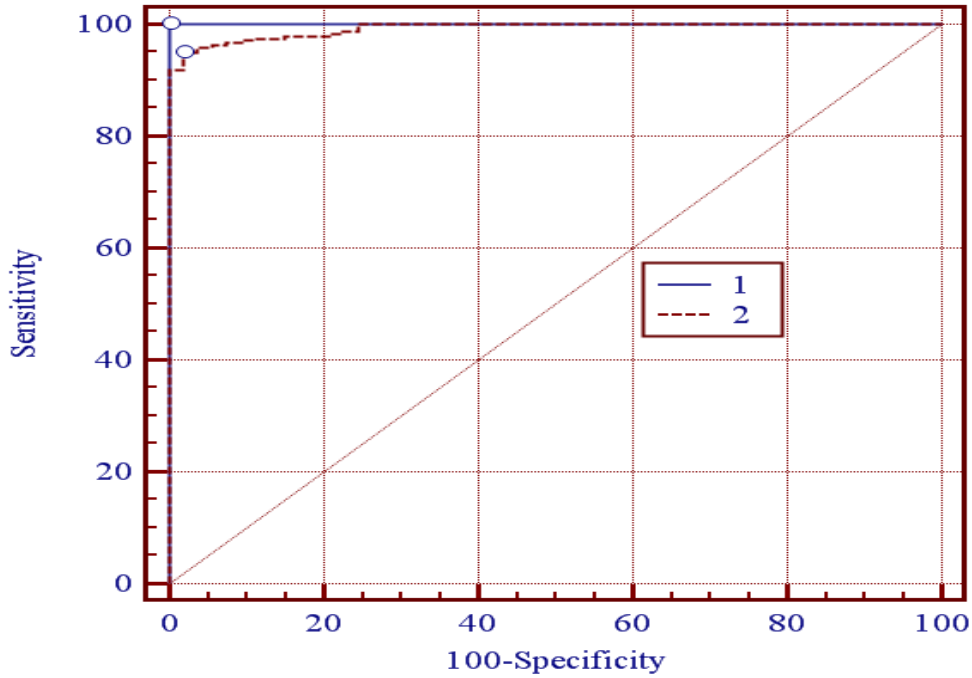

Fig. 1. ROC-curves of models for predicting the risk of development of dysbiosis according to ICPM; 1 – model based on all 58 factor features; 2 – linear neural network model based on four selected factor features

Table 2

Coefficients of the four-factor model for predicting the risk of development of dysbiosis according to ICPM (logistic regression model)

Factor feature	Values of coefficients of the prediction model, b±m	Level of significance of the difference from 0	OR (95% CI of the OR)
X20	-0.62±0.20	0.002*	0.54 (0.36-0.80)
X47	0.19±0.17	0.171	–
X48	-0.55±0.23	0.019*	0.58 (0.37-0.91)
X49	0.27±0.20	0.200	–

Notes: OR– odds ratio; CI – confidence interval

From the analysis of coefficients of the logistic regression model, it was found that the risk of development of dysbiosis according to ICPM with statistical significance (p = 0.002) decreased with increasing levels of content of IL10 in vaginal secretion (OR = 0.54; 95% CI 0.36-0.80) per each unit (pg/ml). There was also a reduction (p = 0.019) in the risk of development of dysbiosis according to ICPM with an increase of content of IL4 in blood (OR = 0.58; 95% CI 0.37-0.91) per each unit (pg/ml).ICPM is the main indicator allowing to

divide patients into groups objectively, i.e. to diagnose normocenosis or grade I or II dysbiosis. Thus, the risk of development of dysbiosis according to ICPM reflects the contribution of significant factor features to the value of the indicator. Construction of a nonlinear neural network model for predicting the risk of development of dysbiosis showed that ICPM can be calculated based on the content in of IL10 (pg/ml) in vaginal secretion, as well as the content of IL2, IL4 and IL6 (pg/ml) in blood. Thus, as already shown for NBI, components of the «interleukin cascade» were included in the model as significant factor features: pro-inflammatory IL2 and IL6 and anti-inflammatory IL4 and IL10. Previously it was found that the levels of all interleukins in vaginal secretions and in the blood had similar dynamics – increase for pro- and decrease for anti-inflammatory, which directly depended on the severity of dysbiosis and was reflected the most at BV.

Conclusions. Results of the study substantiated the assumptions about the pathogenetic role of disregulation of hormonal and immune systems in the occurrence of dysbiosis and BV. The level of significance of differencefrom 0 for coefficients of the logistic regression model for factor traits X47 (IL2 level in blood) and X49 (IL6 level in blood) was statistically low ($p = 0.171$ and 0.200 respectively). It is interesting, that these indicators had positive values of the coefficients of the predicting model, i.e. increased the ICPM. Coefficients of factors features X20 (IL10content in vaginal secretion) and X48 (IL4content in blood) had negative signs, i.e. their high values contributed to the reduction of ICPM. With progression of dysbiosis, these indicators were significantly reduced -4.1 times and 5.5 times respectively ($p < 0.001$). In our opinion, inclusion of these indicators reflected the formation of immunodeficiency, both at local and systemic levels.

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