

The Microbiomic Metaproteome of the Taiga Tick *Ixodes persulcatus* from the Tyumen Region

A. S. Kozlova^{1,2}, A. V. Zgoda^{2*}, N. A. Petushkova², N. A. Bolochkov², V. G. Zgoda², M. A. Salnitska¹, D. V. Kazakov¹, A. V. Lisitsa^{1,2}

¹Institute of Environmental and Agricultural Biology (X-BIO), University of Tyumen, Tyumen, 625003 Russia

²Orekhovich Institute of Biomedical Chemistry, Moscow, 119121 Russia

*E-mail: alex.zgoda23@gmail.com

Received July 07, 2025; in final form, October 28, 2025

DOI: 10.32607/actanaturae.27741

Copyright © 2026 National Research University Higher School of Economics. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

ABSTRACT Metagenomic studies have revealed the taxonomic composition of the taiga tick (*Ixodes persulcatus*) microbiome, whereas metaproteomic data has provided information on the biochemically active fraction of the microbial community residing in the tick. The aim of this study was to characterize the biological processes taking place within the microbiome of the taiga tick *I. persulcatus* using a metaproteomic approach. To expand the range of identifiable proteins, we used two trypsin concentrations in sample preparation for mass spectrometric analysis. The metaproteomes of unfed female and male ticks were analyzed, which enabled identification of protein products encoded by 2,100 genes from microorganisms belonging to 203 bacterial and fungal species. Increased abundance of proteins associated with Ascomycota fungi, particularly abundant in females, were detected. Proteins from the pathogenic *Rickettsia* and *Borrelia* species were identified. These findings enable a transition from a taxonomic metagenomic description to a functional analysis of the microbial consortium role in the physiology of the vector tick, particularly given the identified microbiota differences related to the tick sex.

KEYWORDS taiga tick *Ixodes persulcatus*, microbiome, metaproteome, *Rickettsia*, *Borrelia*, mass spectrometry.

INTRODUCTION

Global climate change causes an expansion of the habitats of many animals, in particular arthropods, and the distribution of related pathogens throughout these newly conquered areas [1]. The fauna of ixodid ticks of the world (Acari: Ixodida) involves approximately 900 species belonging to the families Argasidae, Ixodidae, and Nuttalliellidae [2]. Currently, these ticks are found in various natural zones (from steppes to arctic deserts) and on various animals, from reptiles and birds to mammals [3].

Ixodid ticks (Ixodidae) are key reservoirs and vectors (organism vectors) of human and animal pathogens (tick-borne encephalitis, Crimean hemorrhagic fever, borreliosis, piroplasmiasis, etc.) [4]. Pathogenic tick-borne viruses, bacteria, and fungi have been identified using modern sequencing methods. New pathogenic viruses have been discovered in Europe using metagenomic sequencing [5]. New bacteria of the genus *Rickettsia* have been identified by extensive studies of the tick microbiome [6, 7].

Metaproteomics complements genetic profiling (metagenomic analysis), with the characterization of metabolically active microbial taxa. *Rickettsia* (*Rickettsia* sp.) and *Borrelia* (*Borrelia* sp.) have been identified as key human pathogens transmitted by ticks inhabiting New York City parks [8]. Differences have been found between the metagenomic and metaproteomic profiles of *Ixodes* ticks in North America and Western Europe [8]. Previously, no metaproteomic studies of the taiga tick *I. persulcatus* – widespread in Russia and Asia – had been conducted.

In this study, we attempted to characterize the biological processes taking place in the taiga tick microbiome using metaproteomics.

EXPERIMENTAL

Ixodes persulcatus tick samples

Taiga *I. persulcatus* ticks were collected at the Tyumen State University Biological Station near Lake Kuchak in spring 2024 (April–May) along frequently

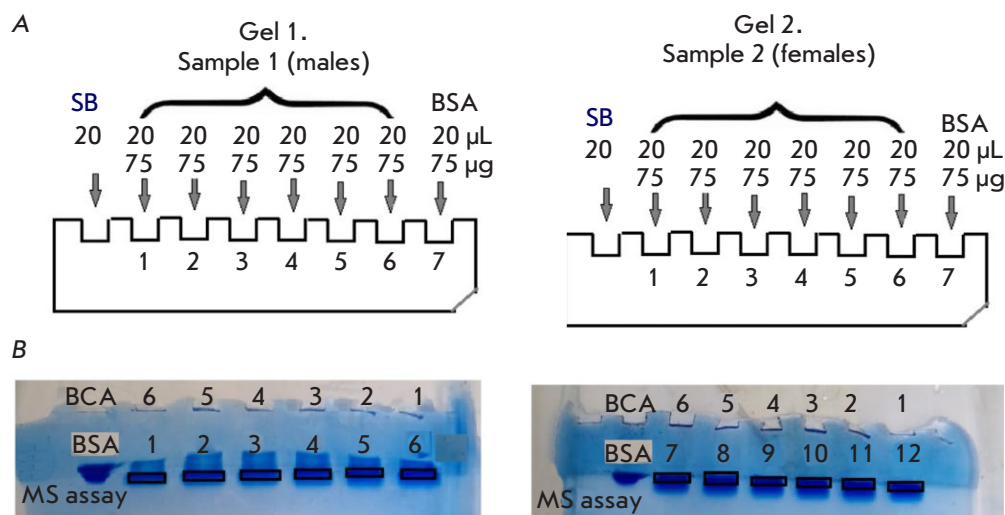


Fig. 1. Loading of protein extracts from *Ixodes persulcatus* tick samples on 1DE gel. (A) SB, electrophoresis buffer, bromophenol blue. (B) Electropherograms of tick protein extracts. MS assay – numbering of technical replicates. BSA – bovine serum albumin, standard

visited nature trails. A 1 m² white flag (a thick white fabric attached to a pole) was used. Ticks caught on the flag were collected with tweezers and transferred to test tubes. After species identification based on morphological criteria, three unfed males and three unfed females were selected. The weight of the three males (15 mg) differed slightly from that of the females (13 mg). For proteomic analysis, samples were stored and transported in liquid nitrogen.

Sample preparation for proteomic profiling

Two pooled samples were used: sample (1) and sample (2) included three male and three female taiga ticks, respectively. Tick bodies (without limbs), frozen at -80°C , were ground with a porcelain pestle in a porcelain mortar, followed by sonication (BANDELIN Sonopuls HD 2070, Berlin, Germany) in an ice bath for two cycles of 50 s each, with a 25 s interval to reduce overheating, and solubilization in the presence of a 2% sodium dodecyl sulfate (SDS) buffer.

The effects of SDS on the inhibition of trypsin enzymatic activity were mitigated by removing the detergent using the 1DE gel concentration procedure (SDS-PAGE without fractionating in resolving gel [9]). The scheme of sample loading on gel is presented in Fig. 1A. Figure 1B shows the resulting electropherograms.

The three most intense bands, each containing approximately 50 µg of protein, were excised from each gel. In-gel digestion was performed using the standard procedure [10]. A tryptic peptide mixture was used for mass spectrometric analysis. Mass spectra of each of the three bands excised from the gels were acquired in three technical replicates. Each of the two samples (males and females) corresponded to nine assays.

Mass spectrometry

Peptides were separated using high-performance liquid chromatography (HPLC, Ultimate 3000 Nano LC System, Thermo Scientific, USA) on a 15 cm C18 column with an internal diameter of 75 µm (Acclaim PepMap RSLC, Thermo Fisher Scientific). The peptides were eluted with a gradient of buffer B (80% acetonitrile, 0.1% formic acid) at a flow rate of 0.3 µL/min. The total chromatography time was 90 min. Measurements were performed on a Q Exactive HF-X Hybrid Quadrupole-Orbitrap mass spectrometer (Thermo Fisher Scientific).

Protein identification

Amino acid sequences of proteins characteristic of the microbial classes most abundant in the microbial communities of ixodid ticks were selected from the UniProtKB database. Four databases were compiled for Alphaproteobacteria, Gammaproteobacteria, Bacillii, and Ascomycota classes containing 36,000, 125,000, 56,000, and 35,000 verified protein-coding genes, respectively.

Peptide/protein identification was performed using the IdentiPROT software (v. 3.2) and the IdentiPy search algorithm [11]. The main identification parameters were as follows: the cleavage enzyme was trypsin; the accuracy of the match between theoretical and experimental peptide weights was ± 5 parts per million, and that of fragment ions was ± 0.01 ; the peptide ion charge state was 2+, 3+, and 4+; the number of possible missed trypsin cleavage sites was no more than one.

Sample preparation details, HPLC-MS/MS modes, and protein identification parameters are provided in the Figshare archive description at <https://doi.org/10.6084/m9.figshare.29469527>. The tsv files con-

taining the metaproteome identification results are available in the [Downloads] tab at the same link.

RESULTS AND DISCUSSION

Identification of proteins from the taiga tick metaproteome

The total number of unique proteins (protein groups) in microorganisms, over all classes, was 1,843, based on data for both female and male samples. If we take more than 276,000 records in UniProtKB as a normalization factor, which means the total number of known bacteria and fungi in potentially expected representatives, then metagenome coverage at the metaproteomic level is approximately 0.01% [8, 12].

On average, the number of identified microbial proteins in the *I. persulcatus* tick in sample groups did not exceed 1,100 (the number of processed mass spectra $n = 9$). For a trypsin : protein ratio of $\approx 1 : 40$, sample 1 (males) contained 482 ± 97 protein identifications and sample 2 (females) contained $1,019 \pm 47$ protein identifications. For a trypsin : protein ratio of $\approx 1 : 100$, sample 1 and sample 2 contained 782 ± 88 and $1,002 \pm 55$ protein identifications, respectively. For a trypsin : protein ratio of $\approx 1 : 40$, the number of identified proteins in the samples of female *I. persulcatus* ticks was 2-fold higher than that in the protein extracts of male ticks. At the same time, at a trypsin : protein ratio of $\approx 1 : 100$, the number of identifications in females and males did not differ significantly and the amount of bacterial and fungal proteins in SDS extracts of male ticks was only 1.3-fold lower than that in the SDS extracts of females. In samples of *I. persulcatus* males, upon addition of a smaller amount of trypsin to a gel fragment containing approximately 50–60 μg of the protein (a trypsin : protein ratio $\approx 1 : 100$), the number of identified microbiome proteins was 1.6-fold higher than that at a trypsin : protein ratio of $\approx 1 : 40$. In samples of *I. persulcatus* females, no similar difference in the number of identified microbiome proteins was found upon varying the trypsin : protein ratio.

Let us consider the results of protein identification of the taiga tick microbiome for nine assays representing three mass spectrometric replicates from each of three gel sections per sample. According to *Table 1*, the number of identifications is distributed depending on the sex of *I. persulcatus* ticks and the trypsin : protein ratio. The highest number of identifications was found for female samples at a lower trypsin concentration (1 : 100). Interpretation of the fragment spectra of 7,008 peptides revealed a total of 1,504 microbial proteins; i.e., 4.65 peptides per protein, on average. This indicator is in good agreement with

Table 1. Number of microbial proteins identified using the IdentiproT proteomic search engine based on nine mass spectra of peptide fragments (3 replicates for each of three gel sections) in a protein extract of the taiga tick *I. persulcatus*

Sample	Trypsin : protein ratio	Number of identifications		
		PSM*	peptides	proteins
No. 1, males	1 : 40	27,709	1,855	1,272
	1 : 100	37,892	2,225	1,427
No. 2, females	1 : 40	46,753	2,509	1,634
	1 : 100	48,896	2,603	1,597

*PSM is the Peptide Spectrum Match that is the number of experimental MS/MS mass spectra statistically significantly matching the theoretical peptide fragmentation spectrum.

the recommendations for assessing the quality of protein identification by mass spectrometric analysis of peptide hydrolysis products [13]. In *Table 1*, the lowest number of identifications (772) corresponds to a protein : trypsin ratio of 1 : 40 in sample 1 for the protein fraction isolated from males. Despite the lower number of identifications, the data quality is comparable to that of the assay described above (females, 1 : 100), because the peptide-to-protein ratio was 4.31.

Figure 2 shows diagrams of the number of identified microbial proteins from *I. persulcatus* ticks, depending on sex and the trypsin : protein ratio. The fraction of proteins common to both trypsin : protein ratio variants in the metaproteomes was 52% and 69% of the total number of identifications in males and females, respectively. It should be noted that the number of metaproteome proteins common to both males and females at the same trypsin : protein ratio was less than 50%. That is, the use of two trypsin concentrations expanded the profile of identified proteins of taiga tick microorganisms.

The heat map in *Fig. 2B* illustrates the quantitative distribution of unique proteins identified in the metaproteomes of female (F) and male (M) *I. persulcatus* ticks under different enzymatic hydrolysis conditions using trypsin at the ratios of 1 : 100 and 1 : 40. Proteins characteristic of five taxonomic classes of microorganisms (Alphaproteobacteria, ArchActBactSpir – a combined category of Archaea, Actinobacteria, Bacteroidia, and Spirochaeta, as well as Ascomycota, Bacilli, and Gammaproteobacteria) are shown. The highest number of class-specific pro-

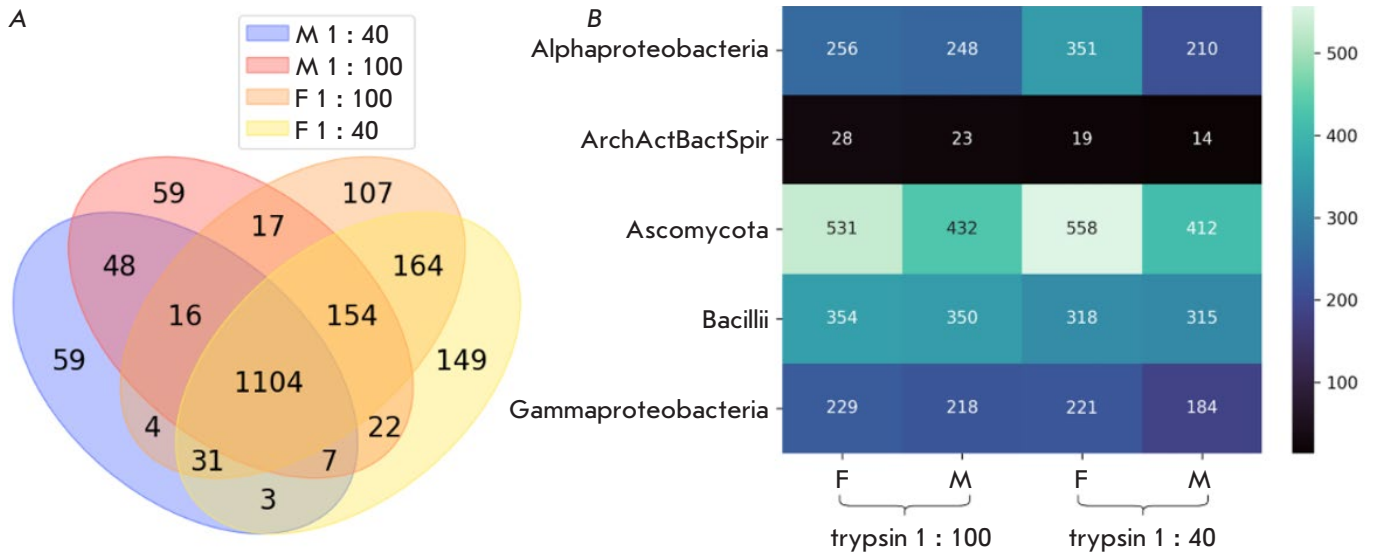


Fig. 2. General information on the metaproteome of unfed males (M) and females (F) of the taiga tick *I. persulcatus*. Identification was performed with the IdentiProt software through databases of the main classes of bacteria and fungi using one or more peptides per protein as a criterion, with two variants of the protein : trypsin concentration ratio: 1 : 100 and 1 : 40. (A) The overlap of sets of identified proteins depending on the tick sex and trypsin concentration. (B) The heat map of the distribution of the number of identified proteins by classes of microorganisms. ArchActBactSpir* is a combination of the classes Archaea, Actinobacteria, Bacteroidia, and Spirochaeta

teins was encountered in members of Ascomycota using two proteolysis protocols, especially in females. Despite the small number of detected actinomycete proteins, those identified by mass spectrometry were present in significant concentrations. This is confirmed by a higher number of identified peptides per protein and high values of the semi-quantitative NSAF estimate.

Microorganisms in the taiga tick metaproteome

Analysis of the taiga tick metaproteome revealed the dominance of fungal proteins: ascomycetes accounted for 61% of all the identified microbial proteins (Fig. 3A–D). Notably, fungal proteins demonstrated exceptionally high identification reliability (more than 8 peptides per protein), whereas bacterial proteins were identified with significantly fewer peptides (less than 7; Fig. 3E,F). This contrast is particularly interesting in light of data [12] showing that pathogenic bacteria predominate in the metaproteome of other ixodid tick species.

We found that trypsin concentration changes did not affect the abundance of fungal proteins, which may be indication of their particular resistance to proteolysis or their high initial levels. The most abundant proteins originated from the single-celled fungi *Neurospora crassa*, *Schizosaccharomyces pombe*, and *Yarrowia lipolytica*. Notably, the semi-quantitative

NSAF indicator showed no significant differences between bacteria and fungi, despite the difference in the number of identified peptides.

Interestingly, the bacterial profile appears to be associated with sexual dimorphism: the bacterial spectrum in males is significantly broader and includes representatives of the *Rhizobium*, *Agrobacterium*, and *Brucella* genera (Fig. 3). Rickettsiae were detected in individuals of both sexes, which is consistent with data [12]. It may be suggested that the fungal component of the microbiome may play a more significant role in the physiology of the taiga tick than previously thought. The prevalence of ascomycete proteins may be considered a factor in adaptation to life on the tick's body surface, whereas bacteria are represented primarily by endosymbionts.

Semi-quantitative composition of the taiga tick metaproteome

Figure 4 shows histograms of the distribution of the NSAF index, which is a relative estimate of the protein content in samples (Fig. 4A,B). Index values decrease exponentially in the range from 0.01 to the maximum protein content. The distribution became more uniform after the exclusion of proteins of the Ascomycota class: the pronounced peak seen in Fig. 4A,B in the region of low intensity values of the semi-quantitative index disappeared (Fig. 4C,D).

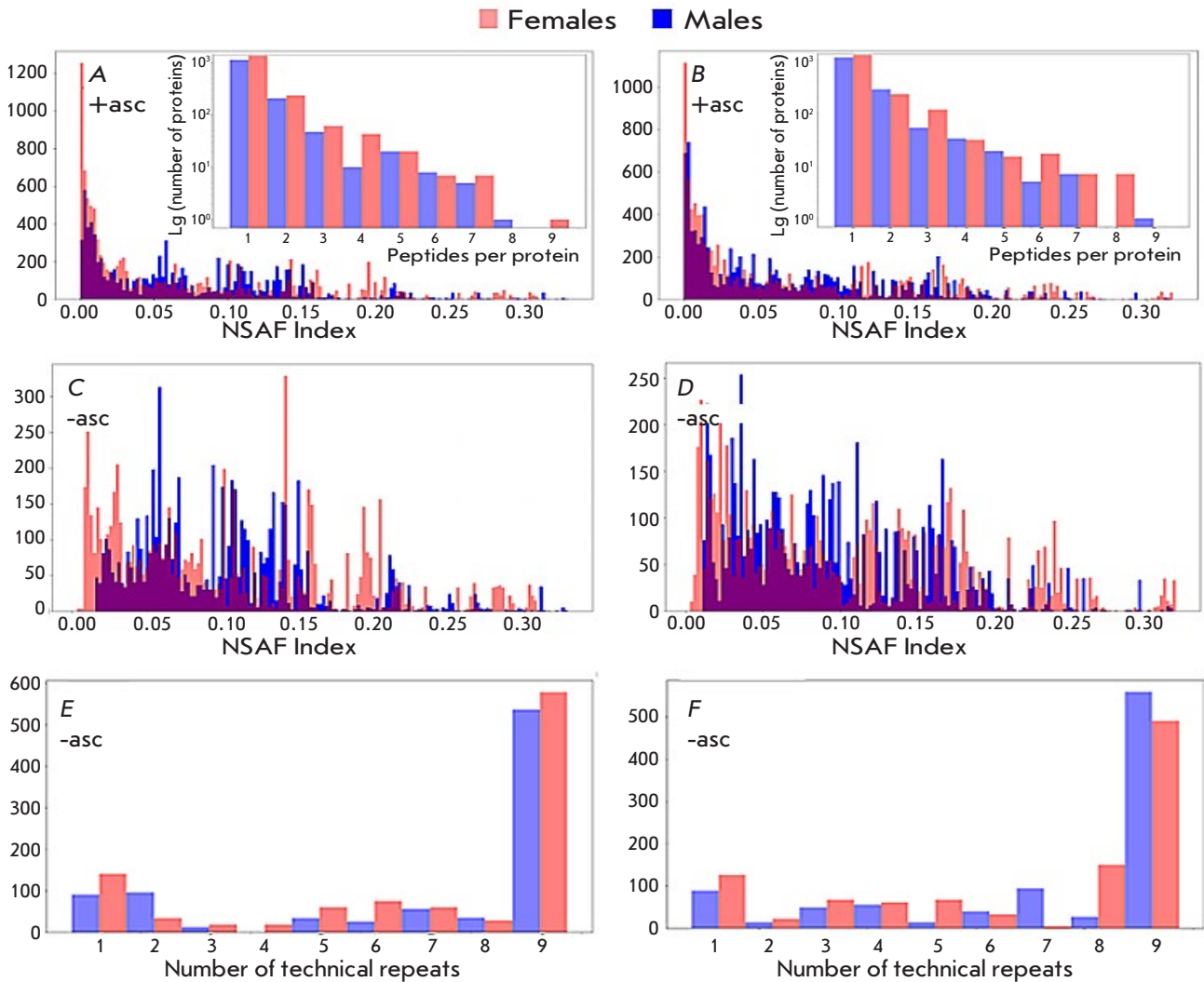


Fig. 4. Protein composition of the taiga tick metaproteome. +/- asc denotes ascomycetes included/excluded from the analysis. Protein : trypsin ratio of 1 : 40 (A, C) and 1 : 100 (B, D). (A, B) Distribution of the semi-quantitative protein content estimate in samples by the NSAF index. (C, D) Number of the mass-spectrometry-detected peptides used to confirm protein identification. (E, F) "Duty cycle" means how often the identification of a protein by one or more peptides was repeated in a series of technical replicates (representatives of the Ascomycota class were excluded from analysis; designated as -asc)

ticks at different amounts of the hydrolytic agent is >1,800 protein products. About a quarter of these are characterized by high identification repeatability in an experiment involving $9 \times 2 = 18$ technical replicates. Information on these reliably identified proteins is shown in *Table 2*.

Table 2 presents the proteins identified in nine technical replicates (three HPLC-MS runs for each of the three gel fragments) in males and females. Metaproteome analysis identified seven housekeeping proteins in each of the nine instrument runs. The core was constituted by energy metabolism enzymes (ATP synthase, malate dehydrogenase, isocitrate de-

hydrogenase) and stress-inducible chaperones (DnaK, HtpG, GroEL).

Interestingly, the synthesis levels of malate dehydrogenase and chaperone GroEL were persistently higher in females. Of particular note was the role of ATP synthase found in 39 bacterial species; ATP synthase subunits accounted for over 60% of the reproducible mass spectrometric identifications. A semi-quantitative assessment (NSAF) confirmed the abundance of ATP synthase (NSAF > 0.20), with minimal values for the chaperone GroEL (NSAF = 0.08). The increased abundance of chaperones in females may reflect an adaptive strategy of the microbiome to

Table 2. Highly abundant bacterial proteins in the metaproteomes of male and female taiga ticks*

No.	Protein	Microorganism	Average NSAF**±SD		Mol. weight, kDa
			Females	Males	
1.	ATP synthase, alpha subunit	<i>Saccharophagus degradans</i> , <i>Teredinibacter turnerae</i> , <i>Dichelobacter nodosus</i> , <i>Francisella tularensis</i> , <i>Pseudomonas putida</i> , etc.	0.22 ± 0.04	0.18 ± 0.05	~55
2.	ATP synthase, beta subunit	<i>Phocaeicola vulgatus</i> , <i>Bacillus cytotoxicus</i> , <i>B. cereus</i> , <i>B. thuringiensis</i> , <i>B. anthracis</i> , etc.	0.21 ± 0.06	0.24 ± 0.04	~52
3.	ATP synthase, beta subunit	<i>Glaesserella parasuis</i>	n/a	0.21 ± 0.05	~55
4.	Chaperone DnaK	<i>Thermosipho africanus</i> , <i>Bartonella henselae</i> , <i>Bartonella tribocorum</i> , <i>Azorhizobium caulinodans</i>	0.18 ± 0.03	0.15 ± 0.05	~65
5.	Chaperone HtpG	<i>Buchnera aphidicola</i> , <i>Rickettsia felis</i> , <i>R. bellii</i> , <i>Rickettsia typhi</i>	0.12 ± 0.04	0.12 ± 0.08	51–72
6.	NADP-dependent isocitrate dehydrogenase	<i>Sphingobium yanoikuyae</i>	0.07 ± 0.01	0.04 ± 0.01	~45
7.	Chaperone GroEL	<i>R. rickettsii</i> , <i>R. prowazekii</i> , <i>R. bellii</i> , <i>R. typhi</i>	n/a	0.08 ± 0.02	~52

*A total protein content to a hydrolytic enzyme (trypsin) ratio of 1 : 100 (millimolar concentrations).

**NSAF is an index showing the amount of protein analyte determined by summing mass spectrometric signals in the absence of an isotope-labeled peptide standard (Normalized Spectral Abundance Factor). n/a – no data, SD – deviation from the mean NSAF index value.

maintain protein homeostasis and ensure the survival of both commensal and pathogenic microorganisms under physiological stress [14]. The ability to maintain viability during host switching, that is, during the transition from a tick to a warm-blooded host, is particularly important for pathogens such as *Borrelia* spp.

Figure 5 presents the results of the functional classification of the proteins in the microbiome metaproteome of male and female *I. persulcatus* ticks. Doughnut charts reflect the relative abundance of proteins involved in biological processes (Gene Ontology : Biological Process, GO : BP) in the metaproteome profile (*Fig. 5A*, males; *Fig. 5B*, females). The analysis revealed significant sex differences in the functional profile of the *I. persulcatus* microbiome. In males, the processes of cell division (15.0%) and ATP synthesis (12.1%) dominate, which indicates the orientation of the microbiota towards maintaining energy-dependent functions and proliferative activity. In females, proteins associated with refolding (22.9%) and cell division (18.6%) predominate, which is related to enhanced protein quality control and proliferative activity. Our data are consistent with the results of [12] emphasizing an increased level of biosynthesis of housekeeping proteins in the metaproteome of the female ticks of another species, *I. ricinus*.

The dendrogram (*Fig. 5C*) reveals functional similarities between biological processes. The dendrogram structure includes clusters combining energy metab-

olism and regulatory mechanisms (glycolysis, one-carbon metabolism, regulation of gene expression), as well as processes maintaining protein homeostasis (protein refolding and translation). An individual cluster summarizes catabolic pathways and cell division, which are coupled with the tricarboxylic acid cycle, apparently reflecting the coordination of energy metabolism and proliferative processes.

CONCLUSION

We studied the metaproteome of *I. persulcatus*, a vector of tick-borne encephalitis and borreliosis in Eurasia, using a metaproteomic approach. Analysis of metaproteomes of the taiga tick *I. persulcatus* required an optimization of the sample preparation procedure, which resulted in a 19% increase in the number of identified proteins using a combined detergent removal approach. We functionally characterized the proteomic composition of the tick microbiome. We moved from taxonomic lists, known from metagenomic data, to the assessment of the biochemical activity of the microbial consortium. In the metaproteome, we found elevated contents of proteins associated with Ascomycota fungi, which was particularly pronounced in females. The identified metaproteome profile, dominated by fungal proteins, differs from data previously reported for other tick species (e.g., *I. scapularis* [8]). Proteins from pathogenic bacteria, rickettsia and borrelia, were observed in the metaproteome. A total of 2,100 microbe-specific proteins were identified, in-

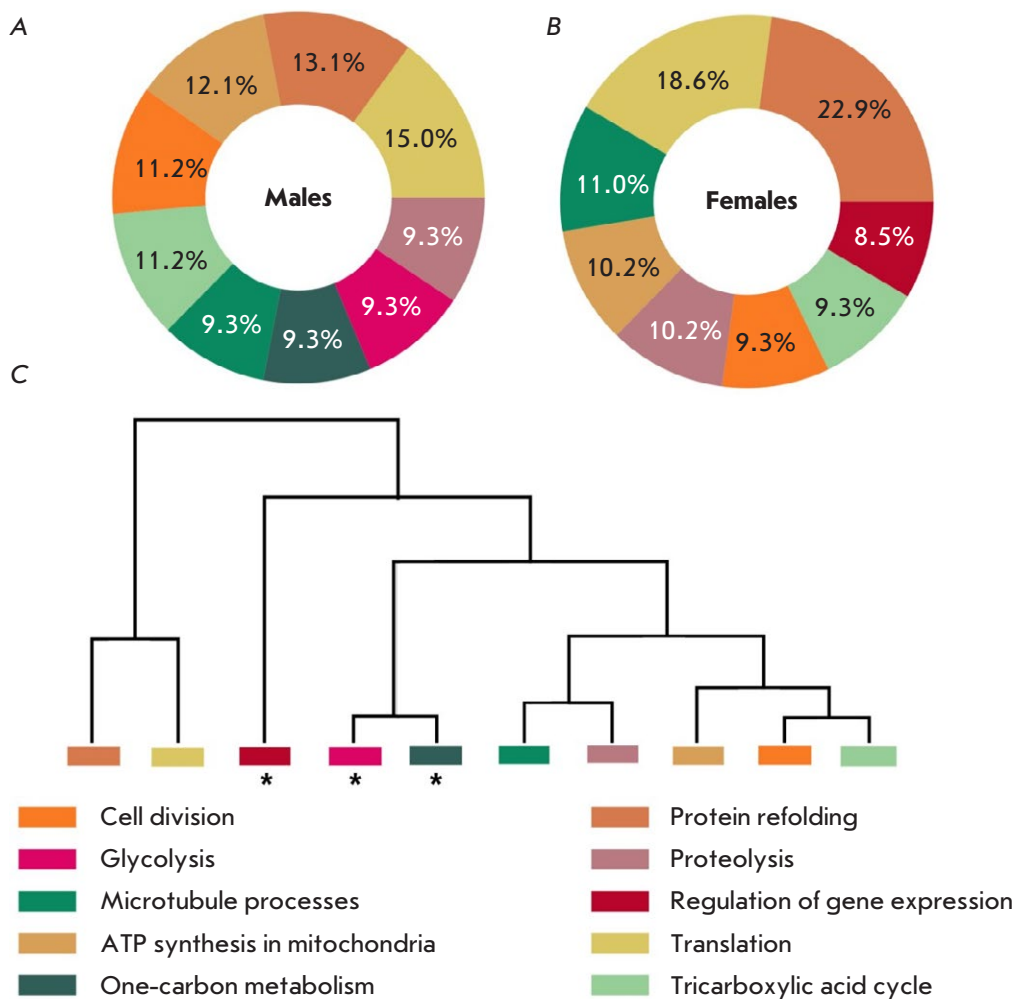


Fig. 5. Protein distribution in the microbiome metaproteome of unfed male (A) and female (B) *Ixodes persulcatus* ticks by biological process categories (Gene Ontology: Biological Process). (C) Clustering of metabolic processes. The * symbol denotes protein groups whose content differs significantly in the microbiome metaproteomes of males and females

dicating the high diversity of the blood-sucking tick microbiota.

It should be considered that low microbial proteome coverage in the setting of a dominant pool of tick host proteins is characteristic of the survey metaproteomics approach for complex samples. This inevitably leads to an underestimation of the fraction of low-abundant, but possibly critically important, pathogens. Their detection requires pre-separation of microbial cells using microfluidics, in combination with targeted proteomic approaches [15].

Therefore, we have been able to identify biochemical processes in the microbiota of the taiga tick *I. persulcatus* (Fig. 5). The obtained data include both bacterial and fungal gene products. The obtained data on the tick microbiota metaproteome are important for developing a multi-omics understanding of *I. persulcatus* activity. ●

The authors thank D.V. Smutin (ITMO National Research University) and A.Kh. Taldaev (Tyumen State University) for providing metadata on microorganisms of Ixodid ticks. Chromatographic-mass spectrometric analysis was performed using equipment of the Human Proteome Core Facility at the V.N. Orekhovich Institute of Biomedical Chemistry. Calculations were performed at the Center for High Performance Computing of the Tyumen State University.

This study was performed within the framework of Agreement No. 075-15-2024-563 of April 24, 2024 “Emergent Biological Threats to Agriculture in Russia and the CIS Countries in the Context of Global Change: Pathogen Identification Using Multi-omics Screening in the Key Vectors and Environment”.

REFERENCES

1. Montero E, González LM, Chaparro A, et al. First record of *Babesia* sp. in Antarctic penguins. *Ticks Tick Borne Dis.* 2016;7(3):498-501. doi: 10.1016/j.ttbdis.2016.02.006
2. Horak IG, Camicas JL, Keirans JE. The Argasidae, Ixodidae and Nuttalliellidae (Acari: Ixodida): A World List of Valid Tick Names. *Exp Appl Acarol.* 2002;28(1-4):27-54. doi: 10.1023/a:1025381712339
3. Beati L, Klompen H. Phylogeography of Ticks (Acari: Ixodida). *Annu Rev Entomol.* 2019;64(1):379-397. doi: 10.1146/annurev-ento-020117-043027
4. Tsapko NV. A Checklist of the ticks (Acari: Ixodidae) of Russia. *Parazitologiya.* 2020;54(4):341-352. doi:10.31857/S1234567806040069
5. Ergunay K, Golubiani G, Kirkitadze G, et al. Ongoing circulation of emerging tick-borne viruses in Poland, Eastern Europe. *PLoS One.* 2025;20(9):e0330544. doi: 10.1371/journal.pone.0330544
6. Polsomboon Nelson S, Ergunay K, Bourke BP, et al. Nanopore-based metagenomics reveal a new *Rickettsia* in Europe. *Ticks Tick Borne Dis.* 2024;15(2):102305. doi: 10.1016/j.ttbdis.2023.102305
7. Ergunay K, Boldbaatar B, Bourke BP, et al. Metagenomic Nanopore Sequencing of Tickborne Pathogens, *Monogolia.* *Emerg Infect Dis.* 2024;30(14):105-110. doi: 10.3201/eid3014.240128
8. Smith HR, Canessa EH, Roy R, Spathis R, Pour MS, Hathout Y. A single tick screening for infectious pathogens using targeted mass spectrometry. *Anal Bioanal Chem.* 2022;414(13):3791-3802. doi: 10.1007/s00216-022-04054-y
9. Shkrigunov T, Pogodin P, Zgoda V, et al. Protocol for Increasing the Sensitivity of MS-Based Protein Detection in Human Chorionic Villi. *Curr Issues Mol Biol.* 2022;44(5):2069-2088. doi: 10.3390/cimb44050140
10. Shevchenko A, Wilm M, Vorm O, Mann M. Mass Spectrometric Sequencing of Proteins from Silver-Stained Polyacrylamide Gels. *Anal Chem.* 1996;68(5):850-858. doi: 10.1021/ac950914h
11. Levitsky LI, Ivanov MV, Lobas AA, et al. IdentiPy: An Extensible Search Engine for Protein Identification in Shotgun Proteomics. *J Proteome Res.* 2018;17(7):2249-2255. doi: 10.1021/acs.jproteome.7b00640
12. Kamburov A, Cavill R, Ebbels TM, Herwig R, Keun HC. Integrated pathway-level analysis of transcriptomics and metabolomics data with IMPaLA. *Bioinformatics.* 2011;27(20):2917-2918. doi: 10.1093/bioinformatics/btr499
13. Paik YK, Omenn GS, Uhlen M, et al. Standard Guidelines for the Chromosome-Centric Human Proteome Project. *J Proteome Res.* 2012;11(4):2005-2013. doi: 10.1021/pr200824a
14. Henderson B, Allan E, Coates AR. Stress Wars: the Direct Role of Host and Bacterial Molecular Chaperones in Bacterial Infection. *Infect Immun.* 2006;74(7):3693-3706. doi: 10.1128/IAI.01882-05
15. Terekhov SS, Smirnov IV, Malakhova MV, et al. Ultra-high-throughput functional profiling of microbiota communities. *Proc Natl Acad Sci U S A.* 2018;115(38):9551-9556. doi: 10.1073/pnas.1811250115