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* MICROBIOLOGICAL STUDY OF AQUATIC ECOSYSTEM OF MONGOLIA

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We have studied microbial ecology of aquatic environments of Mongolia several years randomly, and still our data is not complete till now. Here, we indicated basic microbiological parameters of major lakes and rivers representing Mongolia's aquatic ecosystems. Lakes and rivers water were sampled twice a year, and analyzed for microbiological (total number of bacteria and coliforms, fecal coliforms, enzymatic activity, antibiotic sensitivity and molecular biological analysis) and chemical-physical (ammonia and temperature etc.) parameters. Coliforms were highly correlated with each other and with

ammonia, temperature and pH, suggesting that the simultaneous determination of all variables currently in use in the evaluation of the microbiological quality of waters is probably redundant, and could be simplified, and that coliform should be tested as a sentinel parameter of the microbiological pollution load of lakes and rivers.

Keywords: microorganisms; bacteria; coliforms; aquatic ecology; rivers; pollution of lakes; water quality.

Bacteria occur in all types of surface waters which a natural environment for various groups of organisms, including microorganisms produces, and live in every zones from the surface layer to the bottom deposits. They inhabit clean and heavily polluted waters, fresh inland waters and the salt water of seas, stagnant water and watercourses. Apart from autotrophic forms, one can find heterotrophic and parasitic forms including those that are pathogenic for humans and animals [1].

Microorganisms contribute to the biodegradation and transformation of organic matter, both of autochtonic and allochtonic origin, constituting an important link in the microbial loop, and thus take an active part in the self-purification process of waters [2].

Microbial agents, associated with waterborne outbreaks, include the bacterial organisms *Salmonella*, *Campylobacter* and *Escherichia coli* (*E. coli*) amongst others [3]. These microorganisms may lead to significant health risks in humans, especially infants, the elderly and immune compromised. In severe infections, waterborne diseases may result in chronic illness and even death [4].

Mongolia is not only a water-scarce but also a data-scarce country with regard to environmental information. At the same time, regional effects of global climate change, major land use changes, a booming mining sector, and growing cities with insufficient and decaying water and wastewater infrastructures result in an increasingly unsustainable exploitation and contamination of ground and surface water resources putting at risk both aquatic ecosystems and human health. Presence of water related diseases are evident, when over 50 percent of the population consume water from unimproved sources does not comply with requirements. Therefore it is concluded that the rate of water hygiene and water related diseases is high [5].

The late Professor B. B. Namsaraev and his team from Institute of General and Experimental Biology of Siberian Branch of Russian Academy of Sciences have contributed to lakes and hot springs research.

The aim of this study was to determine the number of bacteria and coliforms occurring in the water of the some rivers and lakes in Mongolia, and to define bacterial pollution as well as some physiological and molecular biological properties.

Materials and methods

Study area. The study areas are located in northwest and northeast of Mongolia. The areas are high in the northwest and low in the northeast with elevation ranging from 602 to 3400 m above the mean sea level. Due to wide application of fertilizer and pesticide in agriculture, industrial and domestic wastes and mining industrial, surface water and soil in the rivers catchment suffer from pollution. The research covered a whole section of the rivers Kharaa, Orkhon, Buyant, Khovd and Onon lying within the

town limits of Darkhan, Erdenet, Khovd and many villages. Kharaa, Orkhon and Onon Rivers are the left bank tributary of the Selenga river; lakes such as Khubsugul, Terkhiin Tsagaan, Ugii Bus belong to the catchment area of the Arctic Ocean drainage basin.

Sampling. Water and biofilm samples were taken for analysis from June, August and September, 2006 to April, 2016 from 69 sampling sites all together, which influenced through livestock, mining industrial and agricultural activities. Water was collected from the middle of the river at a depth of about 20 cm into sterile glass bottles with a capacity of 1 liter, and 100 ml of it was immediately preserved in a solution of formaldehyde (final concentration 0.7%). The collected samples of water were transported to the laboratory in a container and sometimes they were analyzed in the field laboratory.

Enumeration of heterotrophic bacteria. Number of heterotrophic bacteria (CFU) were determined by the spread plate method using Nutrient Agar Medium. The collected samples of water were diluted with sterile distilled water and 0.1 ml was poured into the surface of solid medium. Psychrophilic bacteria were incubated at a temperature of 20° C and the colonies that grew were counted after 3-7 days. Mesophilic bacteria were incubated at a temperature of 37° C and the colonies that grew were counted after 24 h and after 6 days. While colonies of heterotrophic bacteria were calculated per 1 ml of water. All inoculations were conducted in three parallel repeats. After the bacterial colonies were counted, bacterial pure cultures each were transplanted at random each time and after the purity of the culture was checked they were stored in the refrigerator (+4°C) for further tests.

Determination of Coliforms. Total and fecal Coliforms were enumerated by Standard membrane filter technique. With the membrane filter technique appropriate volumes were filtered through Millipore membrane (pore size, 0.45 μ m). Filters were placed directly onto Endo agar, incubating for 24 hr at the 44.5°C, for the Total number of bacteria and fecal Coliforms.

Antibiotic sensitivity of isolated bacteria. The susceptibility of the isolates to various antibiotics were tested by the Disk diffusion method [6]. The following antibiotic discs were used: Gentamicin (10 μ g/ml), Erythromycin (30 μ g/ml), Cefazolin (30 μ g/ml), Ciprofloxacin (30 μ g/ml), Oxacillin (10 μ g/ml), Benzilpenicilline natrium (6 μ g/ml), Tetracycline (30 μ g/ml), Fortum (30 μ g/ml), Nystatin (15 μ g/ml), Claforan (30 μ g/ml) and Chloramphenicol (15 μ g/ml).

Bacterial community by Phylochip analysis. Using a high-density microarray (Phylochip), we examined water column bacterial community DNA extracted from 16 sampling sites of the Kharaa River and its tributaries. The Phylochip microarray probe design approach described was extended and re-applied to all known high-quality 16S rRNA gene sequences containing at least 1,300 nucleotides. Sequences were extracted from the NAST multiple sequence alignment available from the 16S rRNA gene database, greengenes.lbl.gov. This region was selected because it is flanked by universally conversed segments that can be used as PCR priming sites to amplify bacterial or Archaeal genomic material using only 2 to 4 primers. Filtered rRNA gene sequences were clustered to enable selection of perfectly complementary probes representing each sequence of a cluster. Putative amplicons containing 17-mers with

sequence identity to a cluster were included in that cluster. The resulting of the clusters was considered Operational Taxonomic Units (OTUs).

Genomic DNA isolation. Genomic DNA of microbial cells collected on PES membranes were isolated using Phenol-chloroform extraction. The quality and quantity of extracted genomic DNA were determined by a spectrophotometer (NanoDrop 2000 spectrophotometer, Thermo Scientific) using UV absorbance at 230 nm, 260 nm and 280 nm, respectively.

Polymerase chain reaction (PCR) of 16S rRNA gene V3-V4 region. PCR amplification of 16S rRNA gene V3-V4 region was performed before submitting isolated DNA to next generation sequencing (NGS). V3-V4 region of 16S rDNA was targeted by a forward primer (16s_illumina_V3F: 5'-CCTACGGGNGGCWGCAG-3') and a reverse primer (16s_illumina_V4R: 5'-GACTACHVGGGTATCTAATCC-3'). Thermal cycling condition of PCR was: initial denaturation at 95°C for 10 min, 25 cycles with denaturation for 30 s at 95°C, annealing for 1 min at 52°C, extension for 1 min at 72°C, and a final extension at 72°C for 5 min. PCR products were examined by 1.0% of Agarose gel electrophoresis.

Next generation sequencing of microbial 16S rRNA gene V3-V4 region. V3-V4 variable region of sample-barcoded 16S rRNA gene were sequenced with the Illumina MiSeq platform. Raw reads were demultiplexed and paired-end was jointed using "PEAR". Primers were trimmed using "AlienTrimmer" and read quality was filtered with average quality value under 10. Short sequences less than 200 nt were discarded. A software package "Mothur" v.1.33.3 (a SILVA aligned version of the gold database) was used to filter out chimera reads. Effective reads were clustered into OTUs with pairwise distances less than 0.03 cutoff value and each OTU was used for taxonomic assignment. Metagenomics analysis including OTU picking, taxonomic assignment was performed using the software package "Mothur" v.1.33.3 and "QIIME" v1.80 with the Greengenes 16S rRNA Taxonomy Database.

Results

River water. Data concerning the total number of bacteria (TN) and Coliforms (CF) occurring in the water of the five Rivers were presented in Fig. 1.

The total number of microbes on the river Orkhon, Khovd and Buyant were usually around 3×10^4 cells/ml, 1.7×10^4 cells/ml and 1.4×10^4 cells/ml respectively. Coliform bacteria were detected in rivers water 182 cfu/100 ml, 280 cfu/100 ml and 264 cfu/100 ml.

Enumeration of total number of bacteria of the Kharaa River could be stated from the investigation that the Mesophilic total count was relatively high in each of 17 samples. It was over the expected level in the "Moderately polluted" category. The number of Coliform bacteria was the value of the category "Polluted", with the highest number of the Coliform found in sample Kh-B. *E.coli* has been detected in the samples Kh-2 – Kh-7, Kh-D, Kh-E.



Fig. 1. Total number of bacteria and Coliforms

In the Onon River water, the total numbers of microorganisms were ranged between 0.2×10^2 to 0.8×10^2 cells/ml. Pathogenic bacteria such as *Brevudimonas dininita* (97%), *Pseudomonas fluorescens* (99%), *Serratia liquefaciens* (99%) and *Granulicatella elegans* (97%) were detected by semiautomatic test VITEK [®]2.

Physical and chemical parameters were monitored at all sites along the Kharaa River and correlation between total number of bacteria and Coliforms and physical-chemical properties presented in Fig. 2.



Fig. 2. The redundancy analysis correlation of the total number of bacteria, Coliforms and physical - chemical properties of the Kharaa River

As it is in seen Fig. 2 the quantities, amount of nutrients, temperature, conductivity and pH are related to WWTP, Kh-2, and Kh-1 sampling points. The WWTP is located from Darkhan city wastewater treatment plant, Kh-2 and Kh-1ts, which are from Darkhan city wastewater treatment plant to the Kharaa River. Water dissolved oxygen and high concentrations are related to Eroo, Sugnugur and Tunkhel sampling sites. As elevated increases, dissolved oxygen increases and these three points are relatively higher than other sampling points.

Antibiotic suspectibility patterns of bacteria. A total of 219 bacterial isolates (*Staphylococcus spp.* n = 53, *Escherichia coli* n = 18, *Salmonella spp.* n = 64, *Clostridium spp.* n = 40 and *Streptococcus spp.* n = 44) were successfully isolated from Kharaa river and tributaries (Table).

Table

		Samples of spring			Samples of summer			Samples of autumn			
N	Antibiotics	Staph. spp	Salmonella spp.	E.coli	Staph. spp	Salmonella spp.	Strep. spp.	Staph.spp	Cl. perfringens	Salmonella spp.	Strep. spp.
1	Gentamicin	26	32	28	-	32	32	20	24	31	23
2	Erythromycin	-	37	-	-	27	29	20	24	30	29
3	Cefazolin	40	34	20	-	23	23	13	15	30	-
4	Ciprofloxacin	58	-	-	-	-	-	50	22	11	-
5	Oxacillin	-	20	-	-	23	-	17	11	14	-
6	Benzilpenicillin natrium	15	32	27	19	34	26	23	22	21	26
7	Tetracycline	45	33	-	-	33	-	20	23	32	-
8	Fortum	23	23	20	-	28	13	22	17	18	22
9	Nystatin	-	12	-	-	-	-	-	14	13	-
10	Claforan	-	23	21	-	-	22	18	30	-	-
11	Chloramphenicol	31	42	28	-	40	38	30	30	33	-

Antibiotic suspectibility test (inhibition *zone*, mm)

Note: - no inhibition

Staphylococcus spp. isolated from the samples of summer exhibited, the lowest percentage of antibiotic resistance (9.1%), followed by *Streptococcus spp.* (36.4%), *Escherichia coli* (54.5%) and *Staphylococcus spp.* isolated from the samples of spring and *Streptococcus spp.* of summer (63.6%). On the other hand, *Clostridium spp.* showed evidence of highest percentage of antibiotic resistance (100%). The sizes of inhibition zone of the antibiotics of the isolated microorganisms were ranged between 11-58 mm. More than 70% of the bacteria were sensitive to Benzilpenicilline natrium, Gentamicin, Fortum, Cefazolin, Chloramphenicol, Erythromycin, Tetracycline, Claforan, Oxacillin, Ciprofloxacin and Nystatin. Conversely, the highest resistance rate of 72.2% was recorded against Benzilpenicilline natrium.

Bacterial community by Phylochip analysis. For the result of this study, the OTUs represented 79 phyla (classified 73 and unclassified 6), 375 subphyla (classified 194 and unclassified 181), 1704 class (classified 365 and unclassified 1339), 2217 order (classified 252 and unclassified 1965) and 4110 family (classified 274 and unclassified 3836). Phylum *Proteobacteria* dominated for 39,8% and subphylum γ -proteobacteria was occupancy 51,9 %, which showed higher amount of the organic pollution in the Kharaa river water.



Fig. 3. Bacterial community composition of the Kharaa River by top 10 phyla

Distributions of detected Operational taxonomic units (OTUs) at the class level were compared among all sampling sites. The top 10 classes with the highest standard deviations were (in descending order): *Bacteroidales, Betaproteobacteria, Clostridiales, Flavobacteriales, Legionellales, Myxococcales, Peptostreptococcaceae, Rhodobacterales, Saprospirales* and *Sphingomonadales*. Of those classes, only *Betaproteobacteria, Myxococcales* and *Peptostreptococcaceae* had higher relative richness at sampling sites (which absolutely lower of the anthropogenic influence).



Fig. 4. Distribution of relative richness at the Class level.

We focused on the sampling site of the Boroo river (mining area), where 460 OTUs were detected and dominated by of following bacterial Phylums: *Proteobacteria* (39,8%), *Bacteroidetes* (18,11%), *Firmicutes* (13,8%) and *Actinobacteria* (9,7%).



Fig. 5. Constitution of Proteobacteria's classes of Boroo river

Lake water results. Water samples from Ugii, Terkhiin Tsagaan, Khubsugul and Bus Lakes water were assessed. Ugii Lake water contains different number of bacteria in different sampling site ranging from $0.1 \cdot 10^6$ up to $2.0 \cdot 10^6$ and where Gram negative rod shaped bacteria (49%) and Gram positive rod shaped bacteria (36%) dominated. The total number of water bacteria of Terkhiin Tsagaan lake were $0.2 \cdot 10^5 - 1.5 \cdot 10^5$ cells/ml and Khubsugul Lake were $0.1 \cdot 10^4 - 1.2 \cdot 10^5$ cells/ml. Presence of *Lysinibacillus sphaericus* (95%), *Elizabethkingia meningoseptica* (94%), *Kocuria rhizophila* (94%) species in Ugii lake were detected by Semiautomatic test VITEK [®]2.

Results of enumeration of water bacteria showed that Bus Lake water has more bacterial pollution among these lakes (Fig. 6).



Fig. 6. Total number of lake water bacteria Coliforms

The water sample has 10^4 to 10^7 cells/ml, and 10^5 to 10^7 cells/ml in the bottom sediment.

Investigation of enzymatic activity of bacteria of Bus lake are shown in Fig. 7 and 8.



Bacteria with proteolytic activity dominated and the water has 10^7 cells/ml. Cellulolytic bacteria were found in 2–3 times less concentrations in comparison with the previous groups. The water and bottom clay did not exceed 10^6 cells/ml. The total number of amylolytic bacteria were greater than the cellulolytic bacterial group.

Comparative study showned that the bottom sediment has similar total number of aerobic and anaerobic bacteria with cellulolytic and amylolytic activity; where in contrast proteolytic aerobic bacteria dominated. The widespread distribution of these groups may depend on the presence of available substrate concentration (organic pollution).

Genomic DNA isolation from Ugii lake





Universal primer: 27F/341R 315 bp

Fig. 9. Ethidium bromide-stained 0.5% agarose gel of total DNA extracted from bacteria collected from Ugii Lake



Quality of DNA extracted from stool

Universal primer: 8F/1512R 1505 bp



Overall, genomic DNA was higher in concentration and better in quality in samples isolated from lake water than samples from feces. This result, probably due to higher beginning biomass was extracted in water samples (1500 mL) than fecal samples (0.5–1.0 g). In addition, inhibitors in the feces may also interfere the DNA extraction efficiency which was supported by observing low 260/280 and 260/230 ratios in the fecal samples of sheep and cow. Nevertheless, successful PCR amplification of samples from Ugii Lake and feces were both achieved.

Polymerase chain reaction (PCR) of 16S rRNA gene V3-V4 region. The result of PCR amplification of Ugii lake and stool samples were shown in Fig. 11.

Next generation sequencing of microbial 16S rRNA gene V3-V4 region. DNAs of the microbes in fecal samples of cow, horse and sheep were extracted and analyzed by using PCR with primers specific to host-associated microbes. Collected microbial samples from lake water were formalin-fixed, extracted for DNA and analyzed by using PCR-based MST. Fig. 12 shows the principle component of the microbial community composition for the 10 samples (7 lake water, 3 fecal samples). The microbial communities were similar for most lake samples, except for site 7. Site 7, the inflow stream to Ugii Lake, that can have different microbial communities than the lake water. Fecal microbial communities were also away from the lake water groups.



Fig. 11. PCR amplification of Ugii Nuur Lake and feces samples

Note: V3-V4 region (~550 bp) of 16S rRNA gene was targeted. PCR of each sample was conducted in duplicate. M: 100 base pair DNA ladder. H₂O and genomic DNA of *Escherichia coli* (*E. coli*) were used as negative control and positive control, respectively.



Comparison among the bacterial OTUs in 3 different fecal samples. VENNY 2.1 was used to compare individual OTUs from fecal samples. Fig. 13 shows the distinct and overlap OTUs from each sample. There were 8, 16, and 21 distinct OTUs from cow, horse and sheep samples.



Fig. 13. OTU overlaps for fecal samples: cow, sheep (shp), and horse (hrs)

The distinct OTUs from each sample were listed below. The percentages of distinct OTUs among all OTUs in each sample were very low.

8 elements included exclusively in "cow_all": cow_all_specific OTU % in cow
Root;k_Bacteria;p_Bacteroidetes;c_Bacteroidia;o_Bacteroidales;f_p-2534-18B5 0.292%
Root;k_Bacteria;p_Proteobacteria;c_Gammaproteobacteria;o_Aeromonadales;f_Succinivibrionacea
e 0.135%
Root;k_Bacteria;p_Firmicutes;c_Clostridia;o_Clostridiales;f_Peptococcaceae 0.110%
Root;k_Bacteria;p_Bacteroidetes;c_Bacteroidia;o_Bacteroidales;f_[Odoribacteraceae] 0.035%
Root;k_Bacteria;p_Verrucomicrobia;c_Opitutae;o_[Cerasicoccales];f_[Cerasicoccaceae] 0.012%
Root;k_Bacteria;p_Firmicutes;c_Clostridia;o_Clostridiales;f_[Tissierellaceae] 0.009%
Root;k_Bacteria;p_Bacteroidetes;c_[Saprospirae];o_[Saprospirales];unclassified 0.007%
Root;k Bacteria;p Actinobacteria;c Acidimicrobiia;o Acidimicrobiales;unclassified 0.005%
16 elements included exclusively in "hrs all": hrs all specific OTU % in cow
Root;k Bacteria;p Lentisphaerae;c [Lentisphaeria];o Z20;f R4-45B 0.390%
Root;k_Bacteria;p_Fibrobacteres;c_Fibrobacteria;o_Fibrobacterales;f_Fibrobacteraceae 0.178%
Root;k Bacteria;p Spirochaetes;c MVP-15;o PL-11B10;unclassified 0.135%
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 $Root;k_Archaea;p_Euryarchaeota;c_Methanomicrobia;o_Methanomicrobiales;f_Methanocorpuscul aceae 0.095\%$

Root;k_Archaea;p	Euryarchaeota;cThermoplasmata;oE2;f[Methanomassiliicoccaceae] 0.079%
Root;k Bacteria;p	Synergistetes; Synergistia; Synergistales; Synergistaceae 0.076%
Root;k Bacteria;p	Verrucomicrobia; c Verruco-5; o WCHB1-41; f WCHB1-25 0.028%
Root;k Bacteria;p	Firmicutes; cBacilli; oLactobacillales; fLactobacillaceae 0.027%
Root;k Bacteria;p	Firmicutes; Clostridia; Clostridiales; Eubacteriaceae 0.020%
Root;k Bacteria;p	Elusimicrobia; Endomicrobia; unclassified; unclassified 0.018%
Root;k Bacteria;p	Verrucomicrobia; [Pedosphaerae]; [Pedosphaerales]; R4-41B 0.012%
Root;k Bacteria;p	Synergistetes; c Synergistia; o Synergistales; unclassified 0.011%
Root;k Bacteria;p	Synergistetes; Synergistia; Synergistales; Dethiosulfovibrionaceae 0.008%
Root;k Bacteria;p	SR1;unclassified;unclassified;unclassified 0.007%
Root;k_Bacteria;p	Firmicutes;c_Clostridia;o_Clostridiales;f_Dehalobacteriaceae 0.006%
Root;k Bacteria;p	Elusimicrobia;c_Elusimicrobia;o_Elusimicrobiales;f_Elusimicrobiaceae 0.005%
21 elements include	d exclusively in "shp_all": shp_all_specific
Root;k_Bacteria;p	Actinobacteria;cActinobacteria;oActinomycetales;fNocardiaceae 0.145%
Root;k_Bacteria;p	Firmicutes;cBacilli;oLactobacillales;fCarnobacteriaceae 0.101%
Root;k_Bacteria;p	Proteobacteria;cGammaproteobacteria;oPseudomonadales;fPseudomonadace
ae 0.039%	
Root;k_Bacteria;p	Actinobacteria;cActinobacteria;oActinomycetales;fNocardioidaceae 0.034%
Root;k_Bacteria;p	Actinobacteria;cActinobacteria;oActinomycetales;fDietziaceae 0.032%
Root;k_Bacteria;p	_Proteobacteria;c_Gammaproteobacteria;o_Xanthomonadales;f_Xanthomonadace
ae 0.027%	
Root;k_Bacteria;p	Actinobacteria;cActinobacteria;oActinomycetales;fCellulomonadaceae
0.020%	
Root;k_Bacteria;p	_Proteobacteria;c_Alphaproteobacteria;o_Rhizobiales;f_Hyphomicrobiaceae
0.015%	
Root;k_Bacteria;p	_Actinobacteria;c_Actinobacteria;o_Actinomycetales;f_Geodermatophilaceae
0.015%	
Root;k_Bacteria;p	_Actinobacteria;c_Actinobacteria;o_Bifidobacteriales;f_Bifidobacteriaceae
0.014%	
Root;k_Bacteria;p	Actinobacteria;cActinobacteria;oActinomycetales;fBogoriellaceae 0.012%
Root;k_Bacteria;p	Firmicutes;cBacilli;oLactobacillales;fAerococcaceae 0.012%
Root;k_Bacteria;p	Acidobacteria;c[Chloracidobacteria];oRB41;fEllin6075 0.010%
Root;k_Bacteria;p	_Proteobacteria;c_Alphaproteobacteria;o_Rhizobiales;f_Methylobacteriaceae
0.009%	
Root;k_Bacteria;p	Cyanobacteria;cChloroplast;oStramenopiles;unclassified 0.007%
Root;k_Bacteria;p	_Actinobacteria;c_Actinobacteria;o_Actinomycetales;f_Intrasporangiaceae
0.007%	
Root;k_Bacteria;p	_Actinobacteria;c_Actinobacteria;o_Actinomycetales;f_Micromonosporaceae
0.007%	
Root;k_Bacteria;p	_Proteobacteria;c_Alphaproteobacteria;o_Rhodospirillales;f_Rhodospirillaceae
0.006%	
Root;k_Bacteria;p	_Proteobacteria;c_Deltaproteobacteria;o_Myxococcales;f_Polyangiaceae 0.006%
Root;k_Bacteria;p	_Proteobacteria;c_Alphaproteobacteria;o_Rickettsiales;f_mitochondria 0.006%
Root;k_Bacteria;p	Firmicutes;cBacilli;oBacillales;fStaphylococcaceae 0.006%

Comparison of the bacterial OTUs against the fecal sample. Majority of OTUs from each lake water samples (0.01% of total reads or higher) were compared against 3 different fecal samples (0.05% of total reads or higher). Fig. 14 shows the overlap OTUs from Ugii Lake Site U1. There were 22 common OTUs among all three fecal samples and the lake water at U1. And, 20 common OTUs were found compared with microbial communities from cow feces only. Next Fig. 14 showned all the overlapped OTUs (common OTUs) from the cross examination of 7 sampling sites and 3 fecal samples.



Fig. 14. Compare all OTUs from site 1 against all OTUs from three fecal samples

The results showed that all 7 water samples carried the highest common OTUs with cow fecal samples (19-20). This indicates that cows may be the major source of fecal pollution in Ugii Lake.

Conclusion

Scale of pollution caused by bacteria and coliforms of the flowing water judging through different seasons, showed tendency to increase in spring and decrease in autumn. Number of the pathogenic bacteria increased in rivers and lakes depending on the number and scale of the point and non-point pollution sources and their diversity enriches, as well as length of its lifetime extends. The water and environmental sanitation needs of low-income groups in both rural and suburban areas are to be addressed. On average, the MPN, fecal Coliform levels within the rivers and lakes notably exceeded the maximum limit $(1,4x10^3 \text{ cell}/100 \text{ ml})$ of drinking water standard of Mongolia throughout the study period.

According to the investigation among the local people, waterborne disease outbreak usually occurs during the hot and dry season when the water temperature rises, which is optimum for most of the isolated bacterial species (pathogenic bacteria) to grow. Overall, the multiple antibiotic resistance values of the present study indicated that the isolated bacteria received high risk exposure to the tested antibiotics. Therefore, we may conclude that the Kharaa River was contaminated with antibiotic residues. This may be due to overuse and misuse of antibiotics among local people.

Proteolytic bacteria of Bus lake were the major group of bacteria which reached up to 10^9 cell/ml. Cellulolytic and Amylolytic bacteria group were 2-3 times less than proteolytic bacteria group in lake water and bottom sediment. Cellulolytic and Amylolytic bacterial groups have not exceeded 10^6 – 10^7 cell/ml.

Microbial populations were similar in samples collected from the lakeshore and Ugii lake center. And, microbial population composition from the lake intake stands apart from all other lake water samples. At least 15 common bacterial OTUs were found in all water and fecal samples and some of them were host-specific species. It is possible that lake water has been affected by all three types of feces. From the microbial population comparison, cow feces may be the major contamination sources (among three fecal samples tested).

This study provided characterization of bacterial community variability during dry weather in fall season in rivers and lakes catchments. The comparative analysis of water community composition resulted in alternative community-based indicators that could be useful for assessing aquatic ecosystem health of Mongolia.

Results of this investigation showned that our aquatic ecosystem was affected by negative influences of human and natural impacts which lead to deterioration.

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We would like to tribute to our late Professor B. B. Namsaraev and give special acknowledgement for his contribution to study of Water Microbiology of Mongolia, especially on Soda Lake and Hot spring study.

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МИКРОБИОЛОГИЧЕСКОЕ ИССЛЕДОВАНИЕ ВОДНЫХ ЭКОСИСТЕМ МОНГОЛИИ

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Изучение микробной экология водных сред Монголии в течение нескольких лет являлись не систематическими и до сих пор наши данные не являются полными. Мы указали основные микробиологические параметры крупных озер и рек, представляющих водные экосистемы Монголии. Отбор проб воды озер и рек проводились дважды в год и анализировались по микробиологическим (общее количество бактерий и колиформ, фекальных колиформ, ферментативная активность, чувствительность антибиотиков и молекулярно-биологический анализ) и химико-физическим (аммиак и температура и др.) параметры. Формы колиформ были сильно коррелированы друг с другом и с аммиаком, температурой и pH, что свидетельствует о том, что одновременное определение всех переменных, используемых в настоящее время при оценке микробиологического качества вод, вероятно, является избыточным и может быть упрощено, и что колиформа должна быть проверена как часовой параметр микробиологической нагрузки загрязнения озер и рек.

Ключевые слова: микроорганизмы; бактерии; колиформы; водная экология; река; загрязнение озер; качество вод.