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## Impact of flooding on the variability of chemical and microbiological state of chestnut soils of Western Kazakhstan Region

Aliya Nagiyeva<sup>1</sup>, Marat Ongayev<sup>2\*</sup>, Rashit Japarov<sup>3</sup>, Yergali Ayupov<sup>4</sup>, Serik Denizbayev<sup>5</sup>

<sup>1,2,3,4,5</sup>Zhangir Khan West Kazakhstan Agrarian Technical University, Republic of Kazakhstan.

Corresponding author: Marat Ongayev (Email: [marat.onaev@mail.ru](mailto:marat.onaev@mail.ru))

### Abstract

The aim of the research was to investigate changes in the soil microbiome and soil chemicals due to the flooding of agricultural areas, as well as to determine the strength of the relationship between these soil indicators. Deliberate flooding is a reliable way to procure cheap and highly nutritious fodder. More than 400 thousand hectares of flooded lands in the republic are concentrated in Western Kazakhstan, where the research was conducted. The research methodology included, along with an agrochemical study of soil fertility, a metagenomic analysis using high-throughput sequencing. All basic soil parameters of the flooded site were compared with the non-irrigated site, which allowed for the identification of the main changes and processes affecting soil fertility by comparison. The results of the study showed that annual flooding led to an increase in the main soil indicator humus, on average by 0.10–0.28% in genetic horizons in comparison with the non-flooded site. Flooding reduced the alkalinity of the soil solution to 7.7 pH, as well as the degree of salinity to weak. Representatives of bacterial communities Proteobacteria, Actinobacteria, Firmicutes, NA, and Verrucomicrobia dominated in the soil samples of the studied plots, while representatives of Acidobacteria, Bacteroidetes, Planctomycetes, Caldithrix, Chloroflexi, and Tenericutes appeared in smaller shares. A high degree of correlation was found only between the chemical parameters of the soil, pH and the sum of salts, with representatives of the bacterial phylum Verrucomicrobia, NA, Acidobacteria, and Caldithrix. Analysis of the correlation field showed a negative type of relationship between the above indicators.

**Keywords:** Agrochemical indicators, Groundwater, Projective coverage, Sequencing, Flooding, Soil microorganisms, Soil.

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**Transparency:** The authors confirm that the manuscript is an honest, accurate, and transparent account of the study; that no vital features of the study have been omitted; and that any discrepancies from the study as planned have been explained. This study followed all ethical practices during writing.

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## 1. Introduction

Soils are one of the main components of ecosystems, which are most exposed to anthropogenic impact. Currently, there is a sharp increase in soil degradation processes, which leads to the loss of soil biogeocenotic and biospheric functions, as well as a significant reduction in biodiversity. About 30 percent of reclaimed lands in Kazakhstan use spring flooding of agricultural lands. In Western Kazakhstan, agricultural land used for regular flooding occupies more than 70 percent of irrigated land [1]. Deliberate flooding is an important tool for managing agroecological and hydrological processes, affecting the chemical, physical, and biological properties of soil. Studies show that flooding increases organic carbon (Corg) and total nitrogen (Ntot), leading to increased soil fertility [2]. Soil fertility is the main determining property and environment for microorganisms' life activity. Considering soil as a complex system for microorganisms, it should be noted that the composition of bacterial communities of dry-steppe soils is influenced by such factors as organic matter content, pH, moisture, nutrient availability, and temperature [3]. The microbial community is the most sensitive component of soil, primarily responding to anthropogenic impact. Taking into account that soil microorganisms play an exceptional role in the cycle of matter and energy, soil self-purification, productivity of phytocenoses, and sustainability of ecosystems as a whole, one of the priority tasks of modern soil science is to study the transformation of soil microbial communities under anthropogenic impact and their restoration to the initial parameters. Similar results were observed in studies where flooding of organic soils promoted the formation of nitrous oxide hotspots, enhancing denitrification processes (Tomasek et al. [4]). In addition, a study by Rupngam and Messiga [5] observed that flooding increases dissolved organic carbon content, which is associated with anaerobic conditions and decomposition of organic matter. The results of the data obtained from the Amazon floodplain showed that the microbiological metabolic activity of the soil depends more on the type of water than on seasonality and type of land use [6]. Deliberate flooding significantly affects the composition and activity of microbial communities and contributes to changes in the structure of bacterial and archaeal communities by altering salt and organic matter content (Furtak et al. [7]). Gnida et al. [8] note that anaerobic conditions increase the activity of denitrifying bacteria and decrease the activity of microbes responsible for organic matter decomposition. Flooding has a significant effect on the structure of microbial communities. In soils exposed to flooding, total microbial biomass increases, and the proportion of Gram-negative bacteria increases [2]. In addition, changes in the ratio of bacterial and archaeal communities have been observed in zones of periodic reservoir flooding [9]. Soil microbial communities in agroecosystems promote soil health, nutrient cycling, and fertility [10, 11], thereby enhancing pathogen suppression [12, 13] and processes that increase crop yields [14, 15], carbon sequestration through the formation of organic matter bound to minerals [16], and carbon [17, 18]. All of these functions of soil microbes are directly or indirectly related to soil physicochemical properties. Soil is an important reservoir of organic carbon, and prokaryotes are an important component of the decomposition system in soil. Despite the high concentration of organic matter in most soil types, only low concentrations of organic carbon are available to microorganisms. Reasons for this include the conversion of most organic matter originating from plants, animals, and microorganisms to humus through a combination of microbiological and abiotic processes, and the uneven distribution of microorganisms and organic compounds in the soil matrix [19]. The metabolism and survival of soil microorganisms are highly dependent on the availability of water and nutrients. Unlike aquatic environments, soil ecosystem surfaces are subjected to dramatic cyclical changes in water content, from water saturation to extreme drought. A portion of the microbial community is killed in each drying and wetting cycle, resulting in changes in microbial community composition. In addition, the microbial community may change in response to changes in water content and other environmental factors such as pH, oxygen availability, or temperature, which are not yet well understood [20]. The activity of microbial communities increases significantly under anaerobic conditions, which favors accelerated decomposition of organic matter and release of carbon compounds [21]. A certain stock of microorganisms and also the specificity of their distribution in the profile and in time are inherent to each soil type. The taxonomic diversity of the soil microbial community is wide, although generally limited to about 30 different phyla of bacteria. According to the results of studies, the predominant phyla of bacteria include Proteobacteria, Acidobacteria, Actinobacteria, Verrucomicrobia, Bacteroidetes, Chloroflexi, Planctomycetes, Gemmatimonadetes, and Firmicutes, while the rest of the bacteria present in the soil occupy small shares in the total number [22-24]. Ecological niches with elevated salt concentrations and high pH values are habitats for microorganisms with specific physiology enabling them to survive under extreme conditions, leading to a specific set of biochemical mechanisms. Such microbiomes may be important in agriculture as producers of known substances or as targets of fundamentally new bioactive compounds [25]. In addition to high alkalinity, high salinity is also considered lethal to most organisms, but these environments are often inhabited by living organisms and can contain large amounts of biomass of functional and taxonomically diverse communities [26]. Under the influence of irrigation reclamation, the intensity of microflora activity changes: the number of ammonifiers, nitrifiers, and denitrifiers increases, while the number of denitrifiers decreases, and the number of *Azotobacter* increases markedly [27, 28]. In addition, there is an increase in the duration of microbiological activity. Despite the negative factors, namely salinity, soil aridity, and soil moisture deficit, many microorganisms have the property of adapting to extreme factors. The aim of this work was to study the ecological-hydrogeological-reclamation state of the flooded site based on studies of soil indicators, the composition of dominant microbiome taxa, hydrogeological and hydrochemical characteristics of groundwater, and the qualitative composition of herbage. Thus, the article examines soil and microbiological processes in flooded agricultural areas in the arid zone of Western Kazakhstan to determine the changes taking place and the degree of interrelation of the studied soil parameters. As a result, the following two research questions are considered in this study: the ratio of soil microorganisms in flooded and non-flooded areas, as well as the content of the main agrochemical parameters of the soil? What is the degree of relationship between the ratio of the soil microbiome and the content of soil chemical elements? The aim of the research was to investigate changes in the soil microbiome and soil

chemicals due to the flooding of agricultural areas, as well as to determine the strength of the relationship between these soil indicators.

## 2. Materials and Methods

### 2.1. Research Conditions

The research was conducted in 2024 on flooded and non-irrigated experimental plots located on the territory of West Kazakhstan region. The research area is characterized by aridity ( $HTC=0.5-0.3$ ); the sum of positive air temperatures above  $100^{\circ}\text{C}$  fluctuates within 2800-30000  $^{\circ}\text{C}$ ; and the duration of the period with temperatures above  $100^{\circ}\text{C}$  is 155-160 days. During this period, 100-130 mm of precipitation falls, 240-260 mm per year. The frost-free period is 144-155 days. Winter with stable, mainly snow cover is 110-120 days. The research area is characterized by aridity ( $HTC=0.5-0.3$ ); the sum of positive air temperatures above  $100^{\circ}\text{C}$  fluctuates within 2800-30000  $^{\circ}\text{C}$ ; and the duration of the period with temperatures above  $100^{\circ}\text{C}$  is 155-160 days. During this period, 100-130 mm of precipitation falls, 240-260 mm per year. Frost-free period - 144-155 days. Winter with stable snow cover - 110-120 days. The studied soil belongs to chestnut weak-solonaceous heavy loamy soil with high silt content and low humus content. The flat terrain with closed saucer-shaped depressions favoured the concentration of natural flooded agricultural lands with meadow vegetation in the West Kazakhstan Oblast. Flooded agricultural lands of West Kazakhstan oblast are located in the Pre-Caspian lowland, which belongs to the undrained territory due to the absence of groundwater flow gradients. Flooding of agricultural land is carried out annually in the spring period, late March – early April. The duration of flooding before water discharge is, on average, 30 days. Flooding regime and frequency were assessed using in-situ observations and Land-sat 8–9 satellite imagery of the flooded area taken from the USGS website [29]. The NDWI (Normalized Difference Water Index) [29, 30] was used to identify and distinguish open-water or wet-water features in the satellite image against the soil and vegetation background. The Normalized Difference Water Index (NDWI) has been applied to identify water bodies, refine their contours on the map, and track change. NDWI is calculated using a combination of GREEN-NIR (visible green and near infrared). The choice of these wavelengths is based on the maximum reflectance values of water bodies in the green wavelength spectrum and the minimum in the near-infrared spectrum, where vegetation and soil have maximum values. Water reflects virtually no light in the infrared range beyond the visible spectrum, and NDWI effectively utilizes this property to detect and monitor the slightest changes in waterbody content [31-33]. In calculating the NDWI index values, the intervals were taken as a basis and are shown in Table 1. Higher NDWI values close to +1 indicate high water content or the presence of water surfaces (Table 1). While lower values, down to -1, indicate signs of drought [34].

**Table 1.**  
NDWI index interval values.

NDWI Index Values		Type of surface
Low Value	High Values	
0,2	1	water surface
-0,2	0,2	flooded, wet surfaces
-0,3	-0,2	moderately arid, non-water surfaces
-1	-0,3	arid, non-water surfaces

### 3.2. Chemical and Metagenomic Analyses of Soil

For soil chemical analysis, samples were taken from each genetic horizon of the profile, forming pooled samples weighing at least 1 kg. Samples were placed in labeled cloth bags and transported to the laboratory. The soil sample was pre-dried to an air-dry condition, thoroughly pulverized, and sieved through a sieve with a mesh diameter of 1 mm. Chemical analysis of each sample was performed in triplicate. The pH values were measured in a water extract (soil to water ratio 1:5) using a pH meter. The humus content in the soil was determined according to the method of I.V. Turin, which is based on the oxidation of organic matter by a solution of potassium bromate ( $\text{K}_2\text{Cr}_2\text{O}_7$ ) in sulfuric acid ( $\text{H}_2\text{SO}_4$ ) with subsequent quantitative analysis of the amount of reduced chromium on a photoelectrocolorimeter. Nitrate nitrogen was determined by a method that involves extracting nitrates with a 1% potassium aluminum alum solution or a 1 mol/dm<sup>3</sup> (1 N) potassium sulfate solution at a soil sample to solution volume ratio of 1:2.5 and then determining nitrates in the extract using an ion-selective electrode. The maximum values of the total relative error of the method at a two-sided confidence level of  $P = 0.95$  are 30% for a nitrate nitrogen mass fraction in the soil of up to 10 ppm and 20% above 10 ppm. Mobile phosphorus and potassium were determined by the Machigin method (modification of the Central Institute of Agrochemical Service of Agriculture) with extraction using ammonium carbonate (10 g/dm<sup>3</sup>) and subsequent determination of phosphorus on a photoelectrocolorimeter, potassium on a flame photometer. Sulfur content was determined by extracting mobile sulfur from soil with potassium chloride solution, precipitation of sulfates with barium chloride, and their subsequent turbidimetric determination in the form of barium sulfate by optical density of the suspension. Soluble starch was used as a suspension stabilizer. Anion-cation composition in aqueous extract (soil: water 1:5). Anions ( $\text{CO}_3$ ,  $\text{HCO}_3$ ,  $\text{Cl}$ ,  $\text{SO}_4$ ) were determined titrimetrically, argentometrically, and turbidimetrically; cations (Na, K, Ca, Mg) by flame photometry and complexometry [35]. Salinity type was determined according to the table of N.I. Bazilevich and E.N. Pankova. Metagenomic analysis by high-throughput sequencing was carried out to assess microbiome composition. For DNA isolation, 0.2 g of the studied soil samples were taken. The prepared purified DNA preparation was used with universal primers F515 GTGCCAGCMGCCGCGGTAA and R806 GACTACVSGGGTATCTAAT [36] having technology-specific

oligonucleotide adapters. Metagenomic sequencing of the 16S rRNA fragment was performed on a Roche GS Junior pyrosequencer according to the protocol provided by Roche. The data obtained as a result of 16S rRNA gene sequencing were analyzed using the QIIME software [37]. The Illumina MiSeq platform was used to perform microbiological analysis.

### 3. Results and Discussion

To study and compare morphological and chemical indices, two soil transects were laid on the non-flooded and flooded sites. The investigated site was characterized by chestnut-heavy loamy soils. According to the main indicators of soil fertility, there were differences in the content of basic nutritional elements between the non-flooded (control) and flooded plots (Table 2).

**Table 2.**

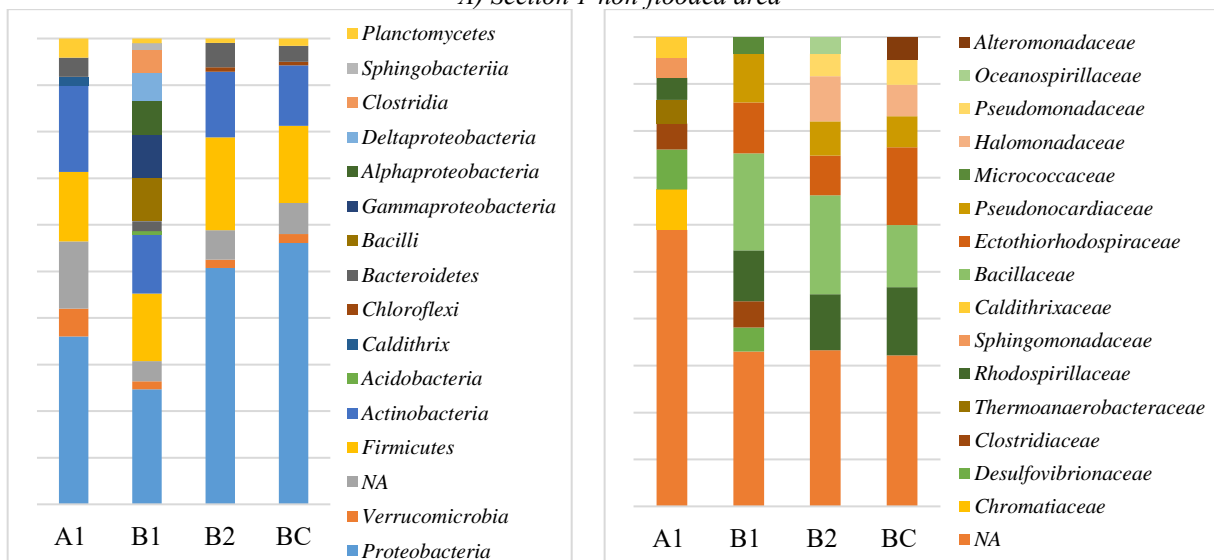
Chemical and microbiological parameters of chestnut soil.

Indicators	Section No.1 (non-flooded section)				
	Horizons				
	A (0–5)	A <sub>1</sub> (5–18)	B <sub>1</sub> (18–30)	B <sub>2</sub> (30–66)	BC (66–120)
pH	turf	7.9	8.5	8.8	9.0
Humus. %		2.27	1.652	1.20	0.67
N-NO <sub>3</sub> . mg/kg		7.28	7.3	7.2	n/o
P <sub>2</sub> O <sub>5</sub> . mg/kg		39.5	58.0	50.6	n/o
K <sub>2</sub> O. mg/kg		319.3	387.0	314.7	n/o
S. mg/kg		5.14	4.47	2.42	n/o
Salinization type		chloride sulphate			
Degree of salinization		medium saline			
<i>Proteobacteria</i>		33.75	-	48.90	54.22
<i>Verrucomicrobia</i>		5.55	-	1.74	1.84
NA		13.51	9.85	6.10	6.44
<i>Firmicutes</i>		13.97	-	19.19	16.04
<i>Actinobacteria</i>		17.31	15.82	13.60	12.50
<i>Acidobacteria</i>		-	-	-	-
<i>Caldithrix</i>		1.83	-	-	-
<i>Chloroflexi</i>		-	-	0.88	0.75
<i>Bacteroidetes</i>		3.81	-	5.03	3.34
<i>Bacilli</i>		-	14.59	-	-
<i>Gammaproteobacteria</i>		-	14.06	-	-
<i>Alphaproteobacteria</i>		-	11.53	-	-
<i>Deltaproteobacteria</i>	-	9.35	-	-	
<i>Clostridia</i>	-	7.28	-	-	
<i>Sphingobacteriia</i>	-	2.27	-	-	
<i>Planctomycetes</i>	-		0.93	1.49	
Section No. 2 (flooded area)					
	A (0–5)	A <sub>1</sub> (5–21)	B <sub>1</sub> (21–31)	B <sub>2</sub> (31–52)	BC (52–110)
pH	turf	7.7	7.8	8.2	8.6
Humus. %		2.37	1.93	1.48	0.32
N-NO <sub>3</sub> . mg/kg		5.27	5.65	5.97	n/o
P <sub>2</sub> O <sub>5</sub> . mg/kg		34.4	35.5	33.3	n/o
K <sub>2</sub> O. mg/kg		274.2	367.74	335.48	n/o
S. mg/kg		5.71	5.25	3.98	n/o
Salinization type		chloride sulphate			sulphate-chloride
Degree of salinization		slightly saline			
<i>Proteobacteria</i>		24.86	24.42	25.88	31.84
<i>Verrucomicrobia</i>		20.51	24.24	18.88	8.04
NA		14.88	13.72	14.61	14.64
<i>Firmicutes</i>		13.21	13.32	16.04	15.25
<i>Actinobacteria</i>		10.20	12.66	14.41	15.07
<i>Acidobacteria</i>		5.11	2.66	1.78	2.00
<i>Caldithrix</i>		2.10	2.30	2.17	2.03
<i>Chloroflexi</i>		1.73	-	-	-
<i>Bacteroidetes</i>		-	1.79	1.65	-

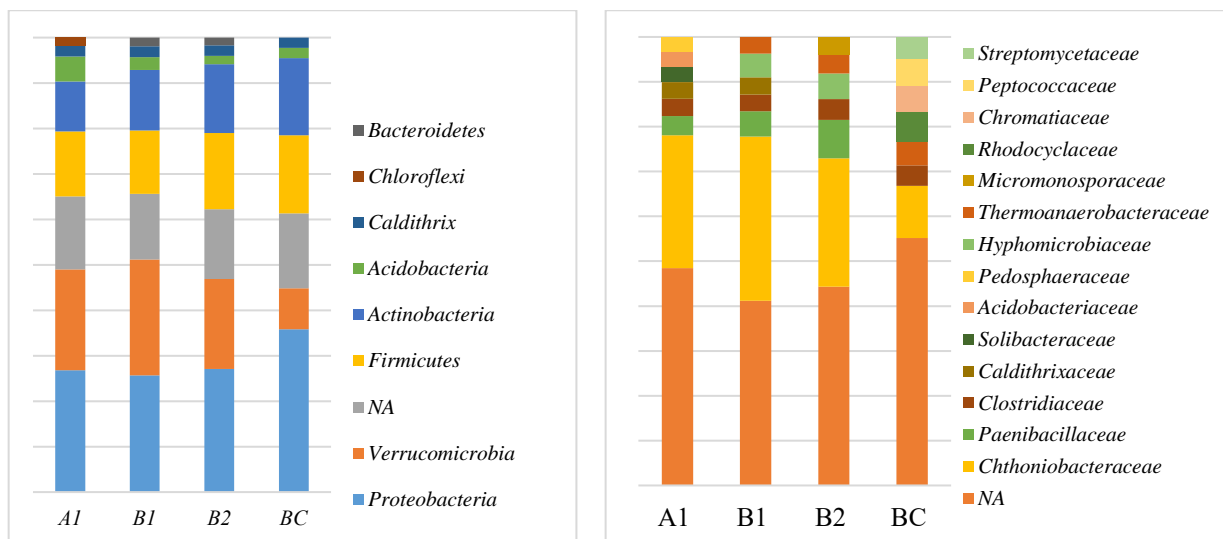
Soil physicochemical properties play a crucial role in determining the composition and function of the soil microbiome [38, 39]. Different soil moisture conditions resulted in distinct microbial community structures [40]. The medium reaction (pH) of the non-flooded section from the surface was noted to be slightly alkaline, which down the profile varied from slightly to strongly alkaline (pH 7.9–9.0); similar dynamics were obtained on the comparative section of the flooded section (pH 7.7–8.6). Studies by scientists show [41–43] that the pH of soil solution is the most dynamic parameter and depends, in addition to the content of hydrogen and aluminum ions, also on a number of factors: changes in climate, environment, aboveground

population, etc. The humus content in the A1 horizon was 2.27% at the control site and 2.37% at the flooded site, which is consistent with the studies of Trost et al. [44] and Kenngott et al. [2], where the soil organic matter content increased similarly with regular flooding. Further down the soil profile, the humus content decreased in both study sites. The nitrate nitrogen content in Section 1 was in the range of 7.28–7.2 mg/kg, with a decrease down the profile, while in Section 2 it was 5.27–5.97 mg/kg, with some increase with depth. Nitrate nitrogen concentration was higher in the non-flooded control plot than in the flooded one, which is explained by denitrification processes and active nitrate removal during periodic soil flooding [4]. Soil properties vary with soil depth [45], which is the case for soil nitrogen [46, 47] and soil phosphorus [48] content. Moreover, soil properties, especially pH, are dynamic and fluctuate with changes in climate [41], environment [42], and aboveground population [43]. There were differences in the content of mobile forms of phosphorus between the study sites. In Section 1, the phosphorus content was 39.5 mg/kg in the A1 horizon, with an increase in the B1 horizon to 58.0 mg/kg and some decrease in the next horizon to 50.6 mg/kg, while in Section 2, this indicator had no significant differences across horizons and was between 34.5–33.3 mg/kg. A number of researchers, Yu et al. [46], Qi et al. [47], and Bai et al. [48] noted a similar trend—the highest mobility of nitrogen and phosphorus and an increase in this property with soil depth [45]. The reactive fractions of phosphorus and nitrogen increased at the beginning of the flooding, mainly through the agricultural use of the upper topography [49]. In terms of potassium content, the non-flooded section had some advantage. In Section 1, the potassium content in A1 and B2 horizons had almost the same values, but in the B1 horizon, there was a slight increase of 387 mg/kg. In Section 2, the upper horizon had lower potassium content (2,74.2 mg/kg) than the underlying B1 and B2 horizons with values of 387.0 and 314.7 mg/kg, respectively. The non-flooded section was characterized by a chloride-sulphate type of salinization and medium salinity throughout the profile. At the flooded site from the surface, the soil had a chloride-sulphate type and a weak degree of salinization; the BC horizon was characterized by sulphate-chloride type with a weak degree of salinization. The results of high-throughput sequencing analysis to determine the dominants of soil microorganisms in the non-flooded and flooded plots revealed two domains - Bacteria (99%), Archaea (up to 1%), as well as unidentifiable microorganisms belonging to bacteria (up to 1%), the presence of viruses was not detected. At the phylum level, 11 taxonomic categories were identified across the study sites, of which the dominant bacterial communities are represented by the phyla *Proteobacteria*, *Actinobacteria*, *Verrucomicrobia*, *NA*, alternating or less frequent representatives of *Bacteroidetes*, *Planctomycetes*, *Acidobacteria*, *Caldithrix*, *Chloroflexi* (Figure 1).

A) Section 1-non-flooded area



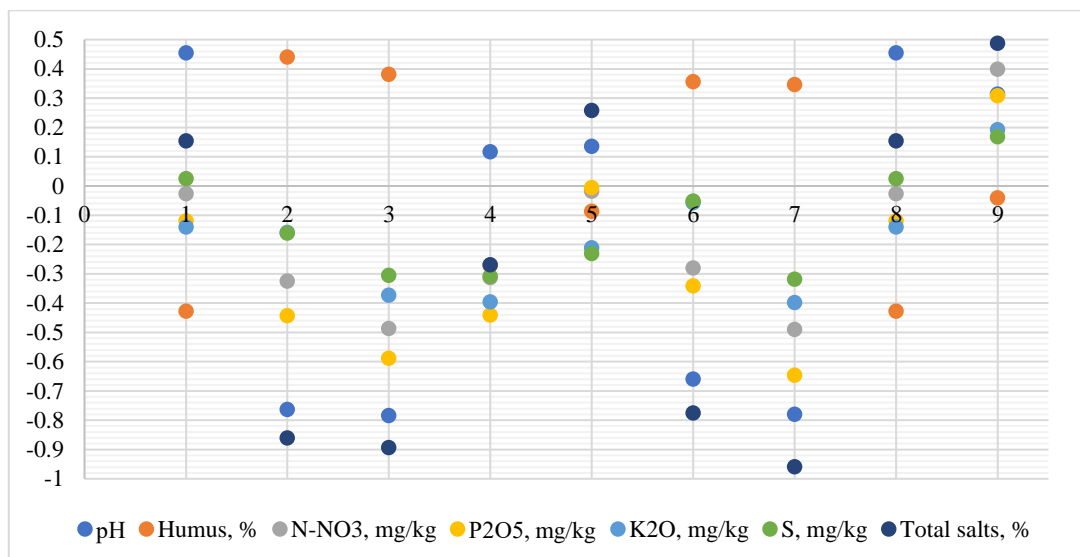
B) Section 2-flooded area



**Figure 1.**

Taxonomic structure of bacterial communities of chestnut soil at phylum and family level, %.

Soil microbial communities are complex and subject to change, which emphasizes the importance of establishing a baseline for comparison with the factors under study. Un-treated controls provide such a baseline to determine whether a change in the soil microbial community is caused by a treatment or an unknown factor [14, 50]. Comparatively investigated sites had differences in both qualitative and quantitative composition. Thus, the content of representatives of *Proteobacteria* communities varied in the profile with a gradual increase to the underlying horizons in the non-flooded and flooded plots, prevailing in the first one (33.7–54.2% and 24.89–31.4%, respectively). *Actinobacteria* representatives at the first site had a variation in the upper fertile horizons and a sharp decrease in the underlying horizons, and accounted for 17.31% in A1 horizon, B1-19.5%, B2 and C horizons-13.6 and 12.5%. Similar distribution by horizons was observed in Firmicutes representatives (13.97–19.19% and 13.21–15.25%, respectively). Representatives of *Gammaproteobacteria*, *Alphaproteobacteria*, *Deltaproteobacteria* are found in horizon B1 with the content of 14.06%, 11.53% and 9.35%, respectively, and representatives of *Verrucomicrobia*, *Bacteroidetes*, *Planctomycetes*, *Acidobacteria*, *Caldithrix*, *Chloroflexi*, *Sphingobacteria* are represented in small amounts or are found only in some horizons. The main mass of unidentifiable bacteria is concentrated in the upper A1 horizon (13.5%) of the non-flooded plot, in the comparison plot they had the same distribution among horizons. At the family level, the comparative analysis on the content of bacterial communities by genetic horizons had some differences. Thus, unidentifiable bacteria (24.89–19.36% and 27.18–24.53%) were leading in the comparative plots in terms of their content in the pro-file. Representatives of the family *Clostridiaceae* were established on the flooded plot in the root-inhabited horizon (2.35%), while on the flooded plot they were located from the surface to the parent rock within 2%. Representatives of the families *Rhodospirillaceae* were found only in the non-flooded area everywhere (2.0–895%). Other representatives of the families *Chromatiaceae* (3.64%), *Desulfovibrionaceae* (3.6–3.1%), *Thermoanaerobacteraceae* (2.11%), *Sphingomonadaceae* (1.84%), *Caldithrixaceae* (1.83%) were found only in the upper A1 horizon, representatives of the families *Bacillaceae* (12.48–8.16%), *Ectothiorhodospiraceae* (6, 52–10.25%), *Pseudonocardaceae* (6.35–4.07%) are found from the B1 horizon to the par-ent rock, families *Micrococcaceae* (2.05%), *Halomonadaceae* (5.59–4.15%), *Pseudomonadaceae* (2.79–3.29%), *Oceanospirillaceae* (2.07%), *Alteromonadaceae* (2.99%) are found in the under-lying horizons. At the flooded site, representatives of the families *Chthoniobacteraceae* (6.52–10.25%), *Paenibacillaceae* (2.37–4.93%), *Clostridiaceae* (2.16–2.70%) are distributed in horizons down the profile, families *Caldithrixaceae* (2.1–2.3%), *Solibacteraceae* (1.93%), *Acidobacteriaceae* (1.87%), *Pedosphaeraceae* (1.85%) were found in the upper A1 horizon, families *Hyphomicrobiaceae* (3.15–3.26%), other representatives of the families *Thermoanaerobacteraceae* (2.2–2.54%), *Micromonosporaceae* (2.27%), *Rhodocyclaceae* (3.22%), *Chromatiaceae* (2.87%), *Peptococcaceae* (2.84%), *Streptomycetaceae* (2.35%) are located in the underlying horizons. Such a distribution of soil communities of microorganisms is explained by the fact that the flooding factor may have had a negative impact on the habitat of the microbiome, which forced them to move to the lower horizons, where it is possible to live more favorably, despite the critical properties - the presence of carbonates, salinity, high density, low pro-vision of organic matter and nutrients, excess or lack of moisture, etc. studies where the authors note that soil microbial communities can be highly susceptible to environmental changes [50-52] and flooding, among other things, has a significant impact on their taxonomic composition [53]. To assess the distribution and movement of microbial communities at the level of phyla depending on the chemical composition of the soil, a correlation analysis was carried out (Figure 2).

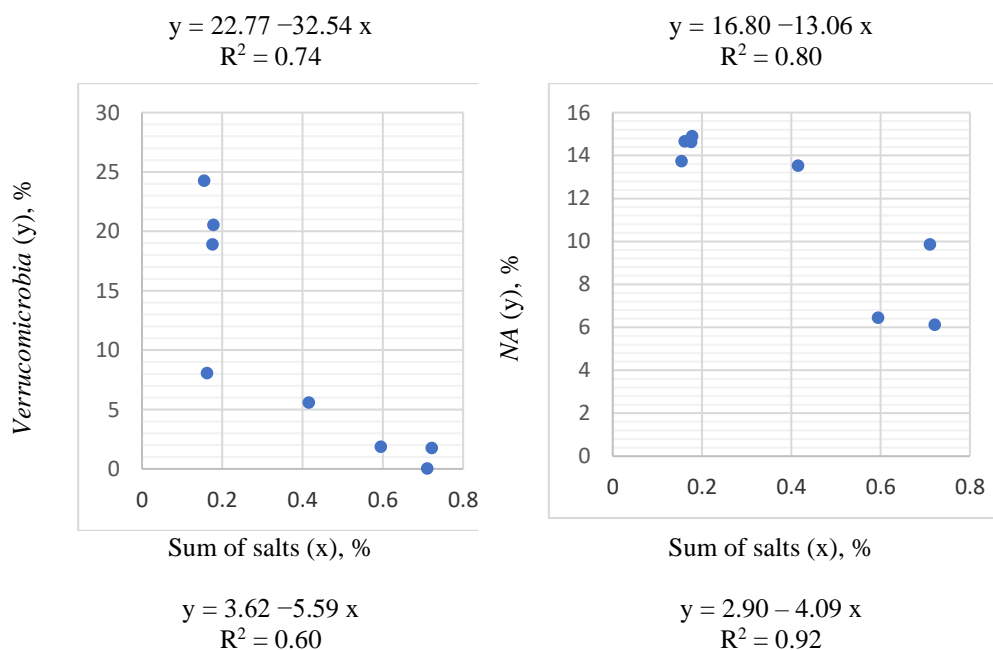


**Figure 2.**

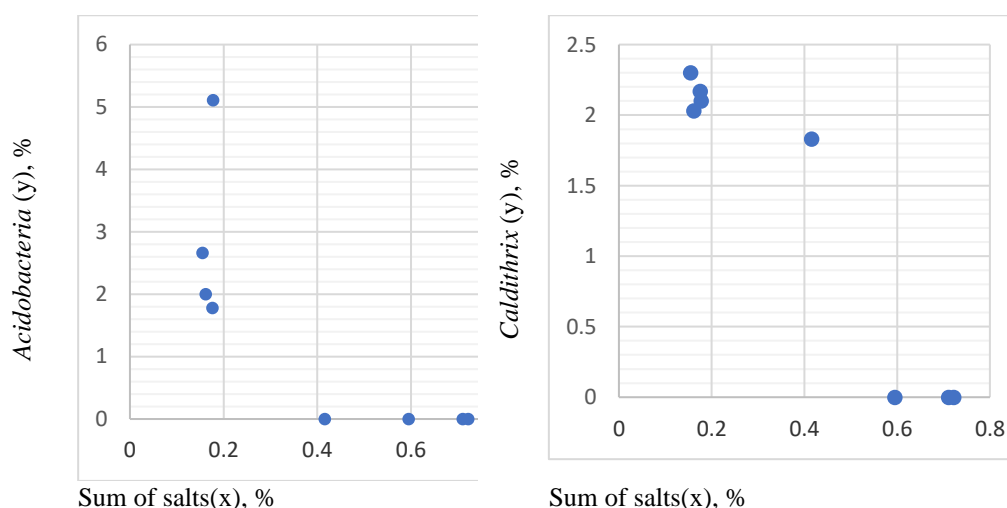
Assessment of the correlation between the indicators of soil chemical composition and the ratio of bacterial communities (1. *Proteobacteria*; 2. *Verrucomicrobia*; 3. *NA*; 4. *Firmicutes*; 5. *Actinobacteria*; 6. *Acidobacteria*; 7. *Caldithrix*; 8. *Chloroflexi*; 9. *Bacteroidetes*).

A high degree of correlation was found between the pH of the soil solution and with representatives of the bacterial phylum *Verrucomicrobia*, *NA*, *Acidobacteria* and *Caldithrix*,  $r = -0.76$ ,  $-0.78$ ,  $-0.66$  and  $0.78$ , respectively. The negative type of relationship showed that with an increase in the alkalinity of the soil solution, the proportion of these groups of bacteria decreased. With other representatives of the groups of soil bacteria, the relationship with the pH of the soil was weak or average. The strongest negative relationship was observed with the indicator of the sum of soil salts. High sensitivity to increased soil salinity was shown by the same representatives of the soil microflora *Verrucomicrobia*, *NA*, *Acidobacteria* and *Caldithrix*,  $-0.86$ ,  $-0.89$ ,  $-0.77$  and  $-0.96$ , respectively.

Since the “sum of soil salts” indicator had a greater range of numerical data coverage than the hydrogen indicator, a regression equation model was built with the first indicator (independent variable) and the soil microbiome (dependent variable). Analysis of the correlation field shows (Figure 3) the presence of a close to straight-line relationship between the indicators of the sum of salts in the soil (x) and the soil microbiome (y), since the activity, ratio and number of soil microorganisms are made dependent on the chemical composition of the soil [38, 39]. Also, soil bacteria have a strong correlation with environmental enzymes, while soil fungi have a strong correlation with nutrients [54].







**Figure 3.** Dependence of the content of soil microbial communities (share of the total number of microorganisms) on the indicator of the sum of soil salts, %.

The coefficient of determination  $R^2$  = from 0.60 in Acidobacteria to 0.92 in Caldithrix shows that 60–92% of the variation in soil microorganisms is explained by the variation of factor x, an indicator of the sum of soil salts, and the remaining 8–40% is due to the action of other factors not included in the model.

#### 4. Conclusion

Thus, the annual flooding led to an increase in the main soil indicator humus, in comparison with the non-flooded area, by an average of 0.10–0.28% (absolute value) in terms of genetic horizons. Flooding contributed to a decrease in the alkalinity of the soil solution to 7.7 pH and the degree of salinity to a weak one (with an average degree of salinity in a non-flooded area). Nitrate content also decreases with depth, while their concentration on non-flooded soils is higher than on flooded soils. The phosphorus content was higher in non-flooded areas, and there were smaller differences when comparing the potassium content in the soil. A comparative analysis of the bacterial community shows a significant difference between the sites. The non-flooded area was dominated by Proteobacteria (33.75–54.22%), Firmicutes (13.97–19.19%), and Actinobacteria (17.31–12.50%), while the flooded area was dominated by Verrucomicrobia (20.51–8.04%) and Acidobacteria (5.11–2.00%), respectively. In the flooded area, the activity of Deltaproteobacteria and Clostridia was manifested in anaerobic conditions, which is typical for the underlying soil horizons. A high degree of correlation was found only between the chemical parameters of the soil—pH and the sum of salts, with representatives of the bacterial phylum Verrucomicrobia, NA, Acidobacteria, and Caldithrix. Analysis of the correlation field showed a negative type of relationship between the above indicators. Flooding of soils has a multifaceted effect on their chemical and microbiological characteristics. Understanding these processes is essential for ecosystem management, agronomic practices, and climate change mitigation.

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