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#### PHD THESIS

## THE FEATURES OF AGROFOREST ECOSYSTEMS AND THEIR IMPACT ON THE ENVIRONMENTAL INDICATORS OF CHUSHANDIAN RESERVOIR'S BUFFER STRIPS

Field of knowledge: 20 - agricultural sciences and food production Specialty: 201 - agronomy

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The dissertation contains the results of own research. The use of ideas, results and texts of other authors have references to the relevant source \_\_\_\_\_ Yan Tengfei

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#### АНОТАЦІЯ

*Янь Тенфей* «Особливості агролісових екосистем та їхній вплив на показники довкілля буферних смуг водосховища Чушандіан ». – Кваліфікаційна наукова праця на правах рукопису. Дисертація на здобуття наукового ступеня доктора філософії за спеціальністю 201 «Агрономія» – Сумський національний аграрний університет, м. Суми, 2023.

**Обґрунтування вибору теми дослідження.** Внесок буферних смуг у глобальну зміну клімату був широко визнаний. Однак управління та використання прибережних смуг залишається проблематичним у всьому світі, особливо у зв'язку зі збільшенням попиту на сільськогосподарські землі. Таким чином, поєднання сільськогосподарського виробництва з перевагами для довкілля стало важливою перешкодою для сталого розвитку прибережних зон (берегу водосховища).

Розвиток агролісових екосистем забезпечує компромісне вирішення цих конфліктів і має великий потенціал для популяризації в майбутньому. Агролісові екосистеми виступають за органічне поєднання лісових дерев або кущів з органічним землеробством або тваринництвом, інтегруючи та синергізуючи переваги надземних та підземних екологічних ніш для максимізації ландшафтних, екологічних та економічних переваг землі. Дослідження характеристик і впливів на довкілля адаптованих агролісових екосистем у прибережних буферних смугах все ще є недостатніми, а знання – обмеженими. Таким чином, посилення досліджень показників довкілля (сукцесії рослинності, структури ґрунту, властивостей ґрунту, екології ґрунту та мікробного співтовариства) та економічної продуктивності (врожайність та економічний дохід) адаптованих агролісових екосистем у смутах все нерохівность та мікробного співтовариства.

прибережних буферних смугах сприяє розвитку та просуванню цього технічного заходу в прибережних зонах. Це також має велике значення для розуміння взаємозв'язку між ґрунтово-рослинним покривом і берегово-водною системою та перевагами для довкілля в екотоні земля-вода.

Наукова новизна одержаних результатів. *Вперше* здійснено комплексний огляд з метою узагальнення стану розвитку та потенціалу застосування існуючих агролісових екосистем у буферних смугах водосховища Чушандіан. *Вперше* досліджено екологічну ефективність таких факторів, як властивості грунту, стабільність ґрунтового агрегату, зміна запасів вуглецю, біорізноманіття рослинності та структура мікробного співтовариства для адаптованих агролісових екосистем у буферних водосховища.

Вперше проаналізовано основні фактори, що впливають на екологічну ефективність адаптованих агролісових екосистем у вказаному регіоні досліджень. Вперше встановлено економічний потенціал цих адаптованих агролісових екосистем. Поглиблено розуміння екологічної ефективності для адаптованих агролісових екосистем (за дрібною шкалою щодо відстані).

Обтрунтовано доцільність створення адаптованих агролісових екосистем у захисних смугах водосховища. *Рекомендовано* заходи щодо підвищення економічної продуктивності та запобіжні заходи для органічного землеробства у адаптованих агролісових екосистемах у буферних смугах водосховища.

**Практичне значення отриманих результатів.** У дисертації наведено вагомі докази доцільності адаптованих агролісових екосистем у буферних смугах водосховища. Завдяки всебічному аналізу літератури було зроблено висновок, що прийняті агролісові екосистеми в буферних смугах водосховища є важливим засобом компромісу між сільськогосподарським виробництвом і екологічними вигодами, який може максимізувати екологічні вигоди при збереженні сільськогосподарської продукції та має великий потенціал для застосування в управлінні та експлуатації прибережної зони в майбутньому.

Автором доведено, що адаптовані агролісові екосистеми в буферній зоні водосховища мають перевагу у збільшенні біорізноманіття рослинності та підтримці популяції видів. Адаптовані агролісові екосистеми в буферних смугах водосховища мають найбільшу кількість видів рослин (27), ніж інші типи землекористування.

Автором встановлено, що адаптовані агролісові екосистеми в буферних смугах водосховища можуть підтримувати життєздатність ґрунту та покращувати якість прибережного середовища існування. Адаптовані агролісові екосистеми в буферних смугах водосховища показали нижчий вміст NH<sub>4</sub>-N (азот амонійний) - 39,55 мг/кг і вищий NO<sub>3</sub>-N (нітрат азоту) - 3,62 мг/кг, ніж інші типи землекористування. Ці екосистеми в буферних смугах водосховища показали вищий вміст MBC (вуглець мікробної біомаси) - 239,52 мг/кг, MBN (азот мікробної біомаси) - 27,75 мг/кг і DOC (розчинений органічний вуглець) - 18,38 мг/кг, ніж інші типи землекористування.

Вміст ТР (загальний фосфор у грунті) - 0,26 г/кг у лісах був нижчим, ніж в інших типах землекористування, що вказує на те, що адаптовані агролісові екосистеми в буферних смугах водосховища мають низьку здатність адсорбувати та утримувати фосфор. BD (об'ємна цільність грунту) - 0,99 г/см<sup>3</sup> і SWC (водомісткість грунту) - 19,28 % у лісистій місцевості були нижчими, ніж в інших типах землекористування, що вказує на те, що адаптовані агролісові екосистеми в буферних смугах водосховища можуть ефективно зменшити вміст води в ґрунті, прискорити інфільтрацію води.

Автором встановлено, що адаптовані агролісові екосистеми в буферних смугах водосховища можуть підтримувати вищу мікробну активність ґрунту, покращувати потенціал запасу вуглецю в ґрунті. Вміст ТС (загальний вуглець) -14,78 г/кг та вміст ТN (загальний азот) - 1,307 г/кг у лісах був вищим, ніж в інших типах землекористування, що вказує на те, що адаптовані агролісові екосистеми в буферних смугах водосховища можуть покращити потенціал поглинання вуглецю в ґрунті.

Індекс Сімпсона, відстань до водотоку та MBN (азот мікробної біомаси) були основними факторами, які впливали на властивості ґрунтів у буферних смугах водойми. Крім того, автором засвідчено, що тип землекористування впливає на властивості ґрунту через стехіометрію ґрунту, а відстань до водотоку впливає на властивості ґрунту через біорізноманіття рослин.

Автором встановлено, що адаптовані агролісові екосистеми в буферних смугах водосховища можуть підтримувати стабільну структуру ґрунту, регулювати зміни запасів вуглецю. МWD (середньозважений діаметр) в агролісових екосистемах був вищим, ніж в інших типах землекористування. Вміст ОСР (пул органічного вуглецю) - 4468,07 г/м<sup>2</sup> у W20 (на ділянці агролісової екосистеми у буферних прибережних смугах резервуара) був найвищим, ніж на всіх інших

ділянках. Розмір частинок мулу, пов'язаний з вуглецем - TC2 може відігравати роль посередника властивості обороту вуглецю.

У дисертації підтверджується, що адаптовані агролісові екосистеми в буферних смугах водосховища можуть покращити мікробне різноманіття, підтримувати стабільну структуру спільноти та посилювати метаболічну активність мікробів. Найпоширенішим типом у буферних смугах водосховища агролісових екосистем є *Proteobacteria*, *Acidobacteriota*, *Actinobacteria* та *Chloroflexi*.

Адаптовані агролісові екосистеми в буферних смугах водосховища показали, що більше ASVs (варіантів послідовності ампліконів) було виснажено, а до 5 найбільших чисельності, класифікованих на рівні типу, належали Acidobacteriota, Actinobacteriota, Proteobacteria, Chloroflexi. Мережа співпоширень показує, що адаптовані агролісові екосистеми мають низький діаметр (5,569) і коефіцієнт кластеризації (5,689), вищими є негативні зв'язки (872).

Функціональна чисельність мікробного співтовариства, рухливість клітин і передача сигналів в екосистемах агролісів були значно нижчими, а метаболізм терпеноїдів, полікетидів, метаболізм вуглеводів, терпеноїдів амінокислот і полікетидів були значно вищими, ніж в інших типах землекористування. Це вказує на те, що адаптовані агролісові екосистеми у буферні смужки резервуару мають сильнішу метаболічну активність для підтримки функціональної стабільності мікробної спільноти.

Основним процесом, який домінував у структурі мікробного співтовариства в агролісових екосистемах, був гомогенний відбір, а потім обмеження розповсюдження. Встановлено, що ТС (загальний вуглець) є головним рушійним фактором, що впливає на структуру мікробного співтовариства в буферних смугах водосховища.

У дисертації показано, що адаптовані агролісові екосистеми в буферних смугах водосховища можуть підтримувати стабільний економічний дохід. Враховуючи екстенсивне управління агролісовими екосистемами у буферних смугах водосховища, автором зазначено, що все ще існує значний потенціал для підвищення врожайності та економічної продуктивності.

Надано пропозиції та питання для уваги керівництва. Автор рекомендує фермерам звернути увагу на посилення управління приводними територіями для покращення якості ґрунту та підтримки продуктивності землі. Крім того, слід суворо заборонити використання фосфоровмісних сполук (добрив, пестицидів тощо) і вдосконалити застосування біологічних заходів контролю, за можливості використовувати органічне землеробство.

Ключові слова: стійке сільське господарство, землекористування, органічне землеробство, зміна клімату, моніторинг, ґрунт, екологія ґрунту, структура ґрунту, текстура ґрунту, вуглецевий цикл, мікробне співтовариство, види рослин, популяція, природні комплекси, зміна запасів вуглецю

#### ANNOTATION

*Yan Tengfei* «The features of agroforest ecosystems and their impact on the environmental indicators of Chushandian reservoir's buffer strips» – Qualifying

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The rational for choosing the research topic. The contribution of buffer zones to global climate change has been widely recognized. However, the management and use of riparian zones remains problematic worldwide, especially as demand for agricultural land increases. Thus, the combination of agricultural production with environmental benefits has become an important obstacle for the sustainable development of coastal zones (reservoir banks).

The development of agroforest ecosystems provides a compromise solution to these conflicts and has great potential for popularization in the future. Agroforest ecosystems advocate the harmonious combination of forest trees or shrubs with organic agriculture or livestock, integrating and synergizing the benefits of above- and belowground ecological niches to maximize the landscape, ecological, and economic benefits of the land.

Research on the characteristics and environmental impacts of adapted agroforest ecosystems in the riparian buffer strips is still insufficient and knowledge is limited. Thus, strengthening research on environmental indicators (vegetation succession, soil structure, soil properties, soil ecology, and microbial community) and economic productivity (yield and economic income) of adapted agroforest ecosystems in the riparian buffer strips contributes to the development and promotion of this technical measure in the riparian zones. It is also of great importance for understanding the relationship between "soil-vegetation cover" and "bank-water system" and the environmental benefits of the "land-water ecotone".

The scientific novelty of the obtained results. For the first time, a comprehensive survey was carried out in order to generalize the state of development and the application potential of the existing agroforest ecosystems in the buffer zones of the Chushandian reservoir. For the first time, the ecological effectiveness of such factors as soil properties, soil aggregate stability, changes in carbon stocks, vegetation biodiversity, and microbial community structure for adapted agroforest ecosystems in reservoir buffer strips was investigated.

*For the first time*, the main factors affecting the ecological efficiency of adapted agroforest ecosystems in the specified research region were analyzed. The economic potential of these adapted agroforest ecosystems has been established *for the first time*. *Improved understanding* of ecological efficiency for adapted agroforest ecosystems (at a fine scale relative to distance).

The expediency of creating adapted agroforest ecosystems in the protective strips of the reservoir is *substantiated*. Measures to increase economic productivity and preventive measures for organic farming in adapted agroforest ecosystems in the buffer zones of the reservoir are *recommended*.

The practical significance of the obtained results. The thesis provides strong evidence of the feasibility of adapted agroforest ecosystems in the buffer zones of the reservoir. Through a comprehensive literature review, it was concluded that adopted agroforest ecosystems in reservoir buffer zones are an important trade-off between agricultural production and environmental benefits that can maximize environmental benefits while preserving agricultural production and have great potential for application in future riparian zone management and operation.

The author proved that adapted agroforest ecosystems in the buffer zone of the reservoir have an advantage in increasing the biodiversity of vegetation and maintaining species population. Adapted agroforestry ecosystems in the buffer strips of the reservoir have the highest number of plant species (27) than other land use types.

The author established that adopted agroforest ecosystems in the reservoir's buffer strips can maintain soil vitality and improve the riparian habitat quality. Adopted agroforest ecosystems in reservoir's buffer strips showed lower NH<sub>4</sub>-N (ammonium nitrogen) content - 39.55 mg/kg and higher NO<sub>3</sub>-N (ammonium nitrogen) - 3.62 mg/kg than the other land-use types. These ecosystems in reservoir's buffer strips showed higher MBC (microbial biomass carbon) content - 239.52 mg/Kg, MBN (microbial biomass nitrogen) content - 27.75 mg/Kg and DOC (dissolved organic carbon) content - 18.38 mg/Kg than the other land use types.

The TP (total phosphorus) content - 0.26g/Kg in woodland was lower than the other land-use types, which indicated that adopted agroforest ecosystems in the reservoir's buffer strips have low capacity to adsorb and retain phosphorous. The BD (bulk density) content - 0.99 g/cm<sup>3</sup> and SWC (soil water) content - 19.28 % in woodland were lower than in other land use types, indicating that adapted agroforest ecosystems in buffer strips of the reservoir can effectively reduce soil water content and accelerate water infiltration.

The author established that adapted agroforest ecosystems in buffer strips of the reservoir can support higher microbial activity of the soil, improve the carbon storage potential in the soil.

The TC (total carbon) content - 14.78 g/Kg and TN (total nitrogen) content - 1.307 g/Kg in woodland was higher than in other land-use types, indicating that adapted agroforest ecosystems in reservoir buffer strips can improve soil carbon sequestration potential.

The Simpson index, distance to the watercourse and MBN (microbial biomass nitrogen) were the main factors that influenced the soil properties in the reservoir's buffer strips. In addition, the author proved that the type of land use affects soil properties through soil stoichiometry, and the distance to a watercourse affects soil properties through plant biodiversity.

The author found that adopted agroforest ecosystems in the reservoir's buffer strips can maintain stable soil structure, regulate carbon stock change.

The MWD (mean weight diameter) in agroforest ecosystems was higher than the other land-use types. The OCP (organic carbon pool) content - 4468.07 g/m<sup>2</sup> in W20 (on the site of the agroforestry ecosystem in the reservoir's buffer strips) was highest than all the other sites. The silt particle- size which was associated with carbon - TC2) may play the role of carbon turnover property intermediary.

The thesis confirms that adapted agroforest ecosystems in reservoir buffer strips can improve microbial diversity, maintain stable community structure, and enhance microbial metabolic activity. The most common type in buffer strips of reservoirs of agroforest ecosystems is *Proteobacteria*, *Acidobacteriota*, *Actinobacteria*, and

#### Chloroflexi.

The adopted agroforest ecosystems in the reservoir's buffer strips showed that more ASVs (amplicon sequence variants) were deplete, and the top 5 abundance classified at Phylum level were *Acidobacteriota*, *Actinobacteriota*, *Proteobacteria*, *Chloroflexi*. The co-occurrence network shows that adopted agroforest ecosystems have low diameter (5.569) and clustering coefficient (5.689), higher negative links (872).

Functional microbial community abundance, cell motility, and signal transduction were significantly lower in agroforest ecosystems, and terpenoid, polyketide, carbohydrate, amino acid terpenoid, and polyketide metabolism were significantly higher than in other land use types. This indicates that the adapted agroforest ecosystems in the buffer strips of the reservoir have stronger metabolic activity to maintain the functional stability of the microbial community.

Homogeneous selection followed by dispersal limitation was the main process that dominated microbial community structure in agroforest ecosystems. It was established that TC (total carbon) is the main driving factor affecting the structure of the microbial community in the reservoir's buffer strips.

The thesis shows that adapted agrofores ecosystems in buffer zones of the reservoir can support stable economic income. Given the extensive management of agroforestry ecosystems in the buffer zones of the reservoir, the author notes that there is still considerable potential for increased yields and economic productivity.

Suggestions and questions for the management's attention are provided. The author recommends that farmers pay attention to strengthening the management of the drive areas to improve the quality of the soil and maintain the productivity of the land. In addition, the use of phosphorus-containing compounds (fertilizers, pesticides, etc.) should be strictly prohibited and the use of biological control measures should be improved, and organic farming should be used whenever possible.

**Keywords:** sustainable agriculture, land use, organic farming, climate change, monitoring, soil, soil ecology, soil structure, soil texture, carbon cycle, microbial community, plant species, population, nature complexes, carbon stock change.

# Articles in scientific publications included in the list of specialized scientific publications of Ukraine

1.**Yan, T.**, Kremenetska, Ye. O., Wan, S., Hu, Q. & He, S. Ecological functions and environmental benefits of reservoir riparian zone. *Bulletin of Sumy National Agrarian University. The series "Agronomy and Biology"*, Volume 2 (44). 2021. P. 73-82. <u>https://doi.org/10.32845/agrobio.2021.2.10</u> (*The applicant particapated in search literatures, conclustion the results, preparation of article for printing*).

2.**Yan, T.**, Kremenetska, Ye. O., Wan, S., Hu, Q. & He, S. Study of community structure and distribution of mixed forest near Nanwan lake. *Bulletin of Sumy National Agrarian University. The series "Agronomy and Biology"*, Volume 3 (45). 2021: 78–86. <u>https://doi.org/10.32845/agrobio.2021.3.10</u> (*The applicant participated in research, experiment design, analysis of the results and writing the article*).

3.**Yan, T.**, Kremenetska, Ye. O., Wan, S., Hu, Q. & He, S. Soil chemical properties and phytodiversity of riparian forest land near Nanwan lake. *Bulletin of Sumy National Agrarian University. The series "Agronomy and Biology"*, Volume 4 (46). 2021: 97-104. <u>https://doi.org/10.32845/agrobio.2021.4.14</u> (*The applicant participated in research, experiment design, analysis of the results and writing the article*).

# Articles in periodical scientific publications indexed in the Web of Science Core Collection and/or Scopus databases

4. **Yan, T.**; Kremenetska, Y.; Zhang, B.; He, S.; Wang, X.; Yu, Z.; Hu, Q.; Liang, X.; Fu, M.; Wang, Z. The Relationship between soil particle size fractions, associated carbon

distribution and physicochemical properties of historical land-use types in newly formed reservoir's buffer strips. *Sustainability*. 2022, 14, 8448.

<u>https://doi.org/10.3390/su14148448</u> (*The applicant participated in research, experiment design, analysis of the results and writing the article*).

5. Yan, T.; Kremenetska, Y.; He, S.; Melnyk, T.; Melnyk, A. The relationship between soil physicochemical properties and vegetation biodiversity in newly formed reservoir's buffer strips. *Applied ecology and environmental research* 2023, 21(4):3153-3175. DOI: <u>http://dx.doi.org/10.15666/aeer/2104\_31533175</u> (*The applicant participated in research, experiment design, analysis of the results and writing the article*).

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#### LIST OF CONDITIONAL ABBREVIATION

- TC Total carbon
- TN Total nitrogen
- **TP** Total phosphorus
- MBC Microbial biomass carbon
- MBN Microbial biomass nitrogen
- DOC Dissolved organic carbon
- NH<sub>4</sub>-N Ammonium nitrogen
- NO<sub>3</sub>-N Nitrate nitrogen
- **BD** Bulk density
- SWC Soil water content
- MWD Mean weight diameter
- OCP Organic carbon pool
- TC1 Sand particle-size associated carbon
- TC2 Silt particle-size associated carbon
- TC3 Clay particle-size associated carbon
- TN1 Sand particle-size associated nitrogen
- TN2 Silt particle-size associated nitrogen
- TN3 Clay particle-size associated nitrogen
- NTI Nearest taxon index
- •Woodland (W) Agroforest ecosystems in the reservoir's buffer strips
- Grassland (G) Grassland in the reservoir's buffer strips
- Cropland (C) Abandoned cropland in the reservoir's buffer strips

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#### **INTRODUCTION**

Actuality of theme. Adopted agroforest ecosystems in buffer strips has been proven to maximize the ecological and economic benefits of riparian zone, and is an important measure for future sustainable agriculture development and climate change. However, it is still widely controversial due to its nature complexes architecture of ecological elements and high-quality requirements for production personnel. It is also worth noting that no study comprehensive monitoring and analysis of the environmental performance (such as plant species and population, soil ecology, soil geochemical process, microbial community et al.,) and economic productivity of adopted agroforest ecosystems in the reservoir's buffer strips, which makes the research data particularly relevant.

**Connection of work with scientific programs, plans, topics.** The dissertation research was conducted within the framework of the project of Scientific Research Plan of Training Techniques for Key Teachers of Xinyang Agriculture and Forestry University "Vegetation succession and soil physicochemical properties in the riparian zone of Chushandian Reservoir" (215003). Part of the research was carried out within the framework of the project of Xinyang Ecological Research Institute Open Fund "Soil carbon sequestration potential and microbial drive mechanism of the typical reservoir's buffer strips in Huai River Catchment''.

The aim and objectives of the study. Adopted agroforest ecosystems in riparian buffer strips has been proven to maximize the ecological and economic benefits of riparian zone, and is an important measure for future sustainable agriculture development. However, the environmental performance and economic productivity of adopted agroforest ecosystems in the riparian buffer strips are not yet well described. Therefore, this study aims to clarify the ecological advantages and economic potential for adopted agroforest ecosystems in the reservoir's buffer strips, and to explore the distribution characteristics and changing patterns of environmental indicators for adopted agroforest ecosystems in the reservoir's buffer strips at fine-scales. To provide significant guidance for deepening understanding of the environmental performance and management measures of adopted agroforest ecosystems in the reservoir's buffer strips.

#### For the purpose were assigned the following tasks:

1.To demonstrate the feasibility of adopted agroforest ecosystems in the reservoir buffer strips.

2.To explore the vegetation succession rule and productivity maintenance mechanism of adopted agroforest ecosystems in the reservoir's buffer strips.

3.To clarify the soil ecological processes and contributions to habitat soil quality of adopted agroforest ecosystems in the reservoir's buffer strips.

4.To elucidate the potential and maintenance mechanism of soil carbon stock change in adopted agroforest ecosystems in the reservoir's buffer strips.

5.To reveal the soil aggregate stability and associated nutrient distribution characteristics of adopted agroforest ecosystems in the reservoir's buffer strips.

6.To illustrate the soil microbial species composition, community structure and ecological process of adopted agroforest ecosystems in the reservoir's buffer strips.

7.To discuss the mechanism of economic productivity maintenance of adopted agroforest ecosystems in the reservoir's buffer strips.

8.To provide recommendations and precautions for agricultural production and ecological management measures of adopted agroforest ecosystems in the reservoir's buffer strips.

**Object of study.** After the impoundment of the Chushandian reservoir, the buffer strips of different land-use types around the shoreline were subject to different degrees of disturbance from flooding and the soil chemical processes were intense. The unique land-use pattern around the reservoir provides ideal research conditions for further study of the characteristic and environmental performance of adopted agroforest ecosystems in the reservoir's buffer strips. This study took the buffer strips of the Chushandian reservoir as the research object, focusing on soil physicochemical properties, vegetation structure, soil aggregates, microbial community, yield, and economic productivity for adopted agroforest ecosystems in the reservoir's buffer strips at fine distance scales.

**Subject of study.** The advantages and potential for adopted agroforest ecosystems in reservoir's buffer strips; The vegetation biodiversity pattern of agroforest ecosytems compare to other land-use types in reservoir's buffer strips; The soil physicochemical properties of agroforest ecosytems compare to other land-use types in reservoir's buffer strips; The soil aggregates stability of adopted agroforest ecosystems compare to other land-use types in the reservoir's buffer strips; The soil microbial community composition and assembly mechanism of adopted agroforest ecosystems compare to other land-use types in reservoir's buffer strips; The yiled and economic productivity of adopted agroforest ecosystems compare to monoculture in the reservoir's buffer strips; Clarified the basic characteristics and environmental performance, as well as the changing patterns of various environmental indicators at fine distance scales for adopted agroforest ecosystems in reservoir's buffer strips, enrich the understanding of agroforest ecosystem in reservoir's buffer strips, and provide scientific theoretical basis for adopted agroforest ecosystems in reservoir's buffer strips.

**Research methods.** Survey and soil sampling, soil physicochemical properties analysis (pH, BD, SWC, TC, TN, TP), soil microbial activity analysis (DOC, MBC, MBN), soil particle-size separation technique, microbiomics (16S amplicon sequencing), Bioinformatics analysis techniques (microbial tax composition and assembly process), statistical (processing of research results), inquiry (yeild and economic income).

The main roadmap of research techniques for this research are as follows (Figure 1.1.):

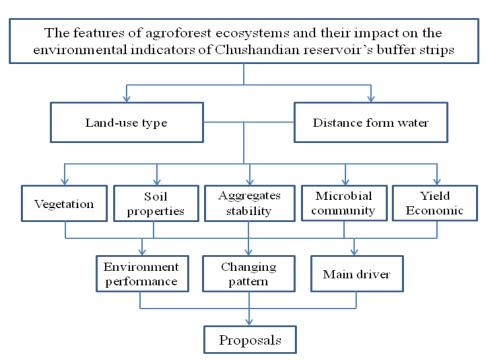


Figure 1.1 The roadmap of research techniques for this study

**Scientific novelty of the obtained results.** The most important scientific results that contain scientific novelty are:

the results obtained for the first time:

 Adopted agroforest ecosystems in the reservoir's buffer strips can still maintain high economic productivity. In the case of strengthening the scientific management and input, there is still considerable potential to increase yeild and economic incomes.

— Proved that adopted agroforest ecosystems in the reservoir's buffer strips can promote carbon sequestration potential, prevent carbon loss, and enhance resistance ability to environmental disturbance.

— Proved that adopted agroforest ecosystems in reservoir's buffer strips have reaative stability microbial community composition and structure, promote microbial metabolic function.

#### there improved:

— Adopted agroforest ecosystems in the reservoir's buffer strips have a low capacity to adsorb and retain phosphorous, which indicates that the farmer needs to be cautions when applying fertilizers and pesticides.

- Adopted agroforest ecosystems in the reservoir's buffer strips have high NH<sub>4</sub>-N and low NO<sub>3</sub>-N content, which indicates that adopted agroforest ecosystems in the reservoir's buffer strips can inhibit the denitrification process, and mitigate greenhouse gas emission.

— Adopted agroforest ecosystems in the reservoir's buffer strips can improve the stability of soil aggregates, and shorten the buffer width setting. The silt particle size associated with carbon content may play a key role in carbon cycle for adopted agroforest ecosystems in the reservoir's buffer strips.

there developed:

Exploring high-value species allocation patterns adapted to adopted agroforest
 ecosystems in the reservoir's buffer strips can improve yeilds and economic incomes.

- Distance to the watercourse induced soil hydraulic gradient was the main factor

influencing the environmental performance of adopted agroforest ecosystems in the reservoir's buffer strips, the farmer needs to pay attention to regional management practice, especially in the near-water area.

The practical significance of the obtained results. We recommend using the materials of the dissertation work when studying the courses "Adaptive farming systems", "Methods and organization of research in agronomy", "Modern problems of agroecology", which are part of the educational and professional program "Agronomy" for masters' training in the field of agroecology, organic production of agricultural products, crop production - on the basis of the Faculty of Agrotechnologies and natural resource management of Sumy NAU. And for the course of "General introduction of agronomy" and "Agroecology" for masters at the Henan Institute of Science and Technology (HIST).

We also recommend that the reservoir management department can use the agroforest ecosystems as a management measure for an eco-economic trade-offs in the reservoir buffer strips.

The main provision of the dissertation was included in the Methodological of modern methods of soil particle-sizes separation, and bioinformatics analysis techniques, for laboratory, practical classes and independent work for master's students of agronomy department from disciplines "edaphology" and "soil microbiology".

**Personal contribution of the applicant.** The author conducted field and laboratory research; an analytical review of the literature was performed; an analysis of the experimental material was carried out; statistical processing of all experimental data and analysis of results was performed. The experimental design, interpretation and

generalization of data, conclusions drawn, and practical suggestions were completed under the guidance of the scientific supervisor.

The recipient's personal contribution to scientific papers published in co-authorship amounted to 50-90%.

Approbation of dissertation results. The main provisions and results of the research were reported and received general scientific approval at the annual scientific seminars and conferences of the teaching staff, postgraduate students, and students of the Sumy National Agrarian University of the Faculty of Agrotechnologies and natural resource management (2019-2023); International scientific conference "The latest scientific achievements in the modern agro-industrial complex", Lublin (2021); International scientific-practical conference "Research of forest and urban ecosystems for sustainable development", Kyiv (2020); III International scientific and practical internet conference "Directions of development of technological systems and logistics in agro-industrial production", Kharkiv (2021); International scientific and practical conference «Honcharivski chytannya», Sumy (2021-2023).

**Publications.** The main results of scientific research were published in 16 scientific articles: 3 articles in specialized publications of Ukraine, 2 articles in journals included in the scientometric databases Scopus and Web of Science, 1 article in a publication of other countries, the rest - in the conference proceedings.

The structure and scope of dissertation. The dissertation consists of an introduction, the main content, which includes 6 chapters, general conclusions, and a list of used literary sources with 237 items. The main content of the work is laid out on 178 pages, including 30 figures and 6 tables. The total volume of work is 186 pages.

#### **CHAPTER 1**

# THE FEATURES AND ENVIRONMENTAL PERFORMANCE OF ADOPTED AGROFOREST ECOSYSTEMS IN RESERVIR BUFFER STRIPS

#### **1.1.** The definition of riparian zone

Before starting the expound, it is necessary to elaborate on the characteristics and definition of the riparian zone so that we can correctly distinguish the connection and distinction between riparian zone and vegetation buffer strips. The vegetation buffer strips are a narrow interface in the riparian zone directly adjacent to the water, while the riparian zone plays an important ecological function as a carrier of the vegetation buffer strips [1-3]. At present, the definition and boundaries of riparian zones are still controversial and there is also a misuse of terms. Commonly used terms include "riparian zone", "water-level fluctuation zone", "buffer zone", "buffer strips", "ecotone zone", "land/inland water ecotones", etc. Many studies even confuse these concepts into one [4-9]. Actually, all of these concepts characterize the water-land interface at different scales, due to the different types of water bodies (rivers, lakes, reservoirs, streams, etc.) and the complexity of the hydrological situation, as well as the objective functions of soils and water bodies, making it difficult to define their scope and boundaries precisely [4]. The size and characteristics of the riparian zone, and whether it is considered a distinct boundary or an interlacing zone, depends on the gradient or rate of change in environmental conditions encountered between the open waterway and the true uplands [10]. Some scholars have defined the definition of riparian zone in terms of the degree of water influence, and in a broad sense, the area directly influenced by water is considered

to be the extent of riparian zone. In a narrow sense, it is considered as the extent of the boundary between waters and land, mainly the area influenced by river flow [11]. Some scholars have also classified riparian zones according to their functions and realistic hydrological operations. For example, riparian zones are horizontally divided into the five-zone model (water zone, non-development zone, special restriction zone, state standard zone A, state standard zone B) in terms of watershed management strategies and non-point source pollution retention purposes [12]. The riparian zone is also divided into Toe Zone, Bank Zone, Overbank Zone, Transitional Zone, and Upland Zone according to the fluctuation pattern of water level. Generally speaking, not all of these 5 parts will appear in the riparian zone, but several will appear. The spatial structure of the riparian zone is a continuous flowing multifunctional linear landscape [7], which exhibits a typical hydraulic gradient pattern along the distance from the watercourse. Correspondingly, the distribution of vegetation on different profiles shows different characteristics, with trees, shrubs, amphibians, and water-holding plants also showing a gradient distribution trend from land to watercourse [13]. With the depth of research, it is gradually recognized that the riparian zone as a complete nature complexes has unique plant and animal community structure and energy flow characteristics. Therefore, the energy flow, material flow, and water cycle of the riparian zone should be considered as a whole ecosystem [10].

#### 1.2. The definition of vegetation buffer strips

The Vegetated buffer zone is defined as a strip of natural vegetation extending a predetermined distance from the water's edge to protect the water quality of a water

supply reservoir and its tributaries by the following processes: 1. Removing sediment, adherents, and dissolved pollutants that reach the buffer strips by providing opportunities for filtration, deposition, infiltration, absorption, adsorption, decomposition, and volatilization; 2. Reducing erosion of riverbeds, banks, and rivers by stabilizing the ground of riverbanks and their tributaries and by reducing overland flow rates; 3. Remove potential sources of non-point source contaminant generation, spills, and illegal dumping from the water's edge [12]. Buffer strips are considered to be the last barrier against pollution from upland agricultural production, and their effectiveness is closely related to the type of vegetation attached to them, which is usually classified into wooded buffer strips, vegetated buffer strips, and zone buffer strips (combining woodland and grassland) according to the configuration pattern of the vegetation [5]. For example, grassland buffer strips are more effective than wooded buffer strips in improving surface roughness, slowing runoff, and increasing infiltration rates. Wooded buffers have a clear advantage over grasslands in terms of carbon stock change and response to climate change [5, 14].

The primary function of the vegetated buffer strips is the retention effectiveness of pollution from agricultural operations in the riparian uplands, which directly affects the amount of surface pollutants that end up into the river, and thus the water quality. The effectiveness of vegetated buffer strips for pollutant mitigation depends on three factors: (i) The physical properties of the buffer strips, such as width, slope, soil type, and vegetation coverage; (ii) The nature of the pollutant in question, such as particle size of the sediment, nitrogen or phosphorus morphology, or biophysical properties of the pesticide (e.g., water solubility and half-life); and (iii) The placement of the buffer strips, such as proximity to the source of pollution. Zhang et al., (2010) suggested that 30m

buffer strips (slope  $\approx 10\%$ ) could remove 80% of the contaminants from upland [15]. Liu et al., (2008) proposed that a 10m width and a 9% slope vegetation buffer strips had the optimal sediment trapping capacity [16]. In contrast, Valkama et al., (2019) argued through meta-analysis that the width of the buffer strips did not significantly affect the retention of nitrogen in surface runoff and groundwater [8]. These differences in buffer strip width setting are mainly caused by the type of pollutant, vegetation placement pattern, and the shelter objective (river, stream, ditch, etc.). In addition, the width of the buffer strips has always been a topical issue in riparian zone protection, especially in areas where agricultural operations are dominant because the width of the buffer strips directly determines the extent of the cultivable area in the riparian zone, representing a trade-off between environmental protection strategies based on ecological benefits and producers based on economic benefits, and a reasonable strips width setting should maximize the environmental benefits of the buffer zone while safeguarding agricultural production [16, 17]. Current studies conducted on buffer strips have focused on nutrient retention above 30m in width, which is clearly beyond what is acceptable to land managers, and fine-scale (1-20m) buffer strips studied are urgently needed to provide more evidence for buffer strip management [18].

#### 1.3. Relationship between vegetation buffer strips and agriculture

In traditional studies, vegetated buffer strips have been used as engineering measures to detain surface source pollution from upland or surrounding agriculture [19, 20]. With the gradual progressive deterioration of the global environment, the ecological footprint of sensitive and fragile riparian zones has also undergone irreversible alteration

[21]. The important role of vegetation buffers in maintaining the diversity of ecological functions in terrestrial-water ecosystems and adjacent agricultural systems has received increasing attention [5, 22]. Over the past century, changes in river and land management practices have degraded the quality and function of riparian ecosystems, simplified the physical structure of riparian habitats, altered river morphology, and reduced water quality [5]. Many scholars believe that agricultural practices are responsible for the degradation of riparian zones [23,24], which undoubtedly exacerbates the conflict between agricultural production and the ecological functions of riparian zones. Riparian farmland provides a unique ecosystem in agricultural landscapes that combines the inherent complexity of riparian ecosystems with the historical lack of agricultural management that has given them rich biodiversity [25, 26]. To guarantee the ecological function of riparian zones, minimum widths of buffer strips are mandatory in many countries, although the range of widths is not uniform, especially in Europe and North America [27]. However, many developing countries in Asia and the tropics still maintain fragmented farming practices, with farmland intertwined with water networks and patchy plots, and pollutants and pesticides discharged from farmland directly into rivers, exacerbating the deterioration of water quality and causing a series of environmental problems [28]. For agricultural producers, the fertile conditions of water and fertilizer in riparian zones are ideal places for carrying out agricultural practices, and the demand for food and increasing population pressure drive the enthusiasm of surrounding farmers for riparian zone land use even though this is not desired for ecological managers. In China, the construction of the Three Gorges Dam inundated large areas of farmland and woodland, creating a 348.9 km<sup>2</sup> floodplain where surrounding farmers cultivate their land during low water periods to compensate for the loss of agricultural opportunities. As a result, a phenomenon of "working with the reservoir floodplain" emerged: water rises and people retreat, water retreats and people cultivate [29]. This contradiction is not irreconcilable in the light of modern sustainable agriculture. Chen et al., (2019) evaluated the ecological benefits of two management practices (conservative and organic farming) in the Three Gorges Reservoir in the reservoir riparian zone and found that organic farming could ensure as many ecological benefits as possible and suggested that more sustainable framing management models should be explored [26].

Although there is no consensus on the definition of sustainable agroecosystems and the scale of application, dimensions, disciplines involved or practical actions, as well as the potential for future applications of sustainable agricultural practices including biofertilizers, agroforest ecosystems, integration of semi-natural landscape elements on fields and farms or management at the landscape scale is not yet sufficient [30]. However, sustainable agricultural practices are still the most effective way to solve future food and environmental problems, especially since the practice patterns that local farmers have developed over time need to be deeply explored and synthesized, and the previous over-reliance on the dissemination of standardized production models has severely limited the application of knowledge generated from farmers' practices [31]. Agroforest ecosystems have been prevalent for centuries as an ancient business model and have been widely demonstrated in production, especially in the context of subsistence agriculture [32]. Adopted agroforest ecosystems in riparian buffer strips have become the primary way to solve the "human-land conflict" in riparian zones.

### **1.4. Agroforest ecosystems**

Agroforest ecosystems have a long history, especially in the tropics, and is seen as an important alternative to slash-and-burn agriculture and cultivation practiced on steep slopes and marginal land [33]. Ecologically based agroforestry practices help restore and maintain biodiversity, bring ecological stability to farms and watersheds, and sustain basic production needs. Agroforest ecosystems advocate the integration of the advantages of organic farming practices and forestry management in a time-series or spatially mixed form, deliberately combining trees and shrubs with crops or livestock to obtain food and economic yeilds while ensuring the ecosystem's potential to defend against climate change [34]. The advantages of agroforest ecosystems are mainly reflected in the rational allocation and utilization of resources and ecological niches in the ecosystem [35-37]. For example, when perennial trees are mixed with annual crops, the tall canopy structure of the trees not only attenuates the range of surface temperature variability and stabilizes the understory microclimate, and their well-developed root systems can also penetrate deep into the ground to absorb deeper groundwater and increase surface moisture through transpiration. The nutrients leached by crops on the surface are also absorbed by the forest root system, improving the efficiency of nutrient utilization [38]. Meanwhile, the rich litter of forest trees can also increase soil fertility and improve soil carbon stock change through microbial decomposition [37, 39].

In fact, the term agroforest ecosystems is a redefinition of traditional agricultural practices. The history of its practice goes back centuries, and many countries have developed locally adapted agroforestry measures [40]. However, it gradually disappeared

with the development of modern machinery and the expansion of agricultural land concentration. It was not until the 1980s, when fossil fuels became increasingly depleted and environmental pressures came to the forefront, that agroforest ecosystems re-entered the scene and were recognized as an important approach to agricultural practices to cope with the increasing extremes of climate change in the future [38]. Although the adoption of agroforestry practices may result in the loss of some yeilds, the resilience advantage shown in the event of extreme climatic occurred remains a very promising development prospect [41]. Agroforest ecosystems are effective in improving surface soil thermodynamic properties, stabilizing soil moisture, increasing water infiltration, reducing soil mineralization rates, increasing soil carbon sequestration potential, accelerating nutrient cycling, and enhancing crop yeilds (planting legumes), among other advantages [3, 42]. Yet, due to the reasons of current standards, configuration mode, policy guidance for agroforest ecosystems, and agricultural technicians not receiving systematic training, farmers are still more inclined to adopt conservative and mature farming systems, but not dare to try agroforestry practices. At the same time, agroforestry practices have excessive requirements for farmers' management level and cognitive ability. Therefore, the implementation of agroforest ecosystems is still a long way off [40, 43].

Riparian vegetation buffer strips are widely recognized as an important component of agroforest ecosystems (include silvoarable and silvopastoral systems, through orchard intercropping, forest farming, riparian buffer strips, and windbreaks) [38, 44], has been widely recognized in developed countries such as Europe and North America [45, 46]. Meanwhile, adopted agroforest ecosystems in riparian buffers have also shown numerous production and environmental advantages. For example, adopted agroforest ecosystems and grassland in riparian buffers can effectively mitigate the loss and increase the sorption properties of antibiotics that came from upland agriculture ecosystems [47]. Adopted agroforest ecosystems in riparian buffer strips have stronger vegetation productivity [1]. Also, the implementation of agroforest ecosystems in riparian buffers can effectively increase the carbon sequestration potential of above-ground trees and below-ground soils, reduce the rate of mineralization, and reduce the emission of greenhouse gases such as CO<sub>2</sub> and N<sub>2</sub>O [2, 34, 39]. Hence, adopting agroforest ecosystems in riparian buffer strips is a promising management practice and nature complexes in terms of environmental benefits and land production synergy. However, due to the management complexity and acceptability of agroforst ecosystems, the understanding of adopted agroforst ecosystems in riparian buffer strips still needs to be deepened.

### 1.5. Environmental performance of vegetation buffer strips

The development of agroforest ecosystems in riparian buffer strips is an important measure for sustainable agriculture and the study of riparian buffers depends mainly on the scales at which we discussed. The nature complexes of riparian conditions, hydrological environment, tourism, agricultural production, animal husbandry, and alluvial flows have created a fragmented habitat in the riparian buffers [48], making it difficult to summarize and elaborate ecological patterns in this region. Current research on vegetation buffer strips has mainly focused on width setting, physical placement of vegetation type, and nutrient retention efficiency [17, 49, 50], while research on the

dynamics of soil physicochemical properties and influencing factors in buffer strips is still lacking. As an important functional component of the riparian zone, buffer strips and buffer zones jointly maintain the stability and ecological balance of the shoreline at different scales and also face common and individual ecological and environmental problems. The buffer strips, as an interactive interface directly adjacent to the watercourse, has always been a hotspot for various soil geochemical processes and is influenced by both abiotic factors from the watercourse and upland [27, 51, 52]. In addition, soil properties in buffer strips are the result of a combination of chemical processes, environmental factors, vegetation types, and litter input and output. For example, DOC, MBC, and MBN characterize labile components of the soil nutrient pool with rapid turnover and are important indicators of microbial activity in the ecosystem [53, 54]. Soil stoichiometric ratio can regulate nutrient cycling through microbial activity and mineralization rates [55, 56]. Likewise, soil texture has been widely shown to exhibit a close relationship with soil nutrient distribution [57, 58]. Vegetation modulates the content and ratio of soil nutrients mainly through the quantity and quality of above-ground litter inputs and below-ground root exudates dynamically [59]. Hence, the study of the relationship between soil physicochemical properties and vegetation biodiversity in buffer strips can help scientists better understand the formation process and structural functions of soils and can provide references for studying ecology proecess such as soil-plant relationship, spatial patterns of vegetation, soil erosion, soil ecology and land use changes [58, 60].

# 1.5.1. Vegetation

The vegetation on riparian buffer strips plays an important role as a cushion in the whole ecosystem and is a key chain in the ecosystem cycle [61, 62]. Among them, hydrological conditions are the core factors in the formation, modification, and succession of vegetation buffer strips. The lakes act on the shoreline through physical effects such as water impact, creating different riparian zone habitats. Propagules of different plants spread with hydrological movements. Fragmented riparian zone habitats form different plant community structures and pioneer species. Likewise, the distribution of plant communities after formation in turn acts on the physicochemical processes of hydrology [63-65]. The most distinctive feature of the riparian buffer strips is the difference in density and species composition from adjacent terrestrial regions [24].

The vegetation has a typical hydrophilic nature, which is the result of the action of environmental conditions in the riparian zone. Specifically, the soil has high year-round water availability and the roots have continuous access to water due to the relatively high water table. In addition, these areas are subject to frequent flooding, so hydrophilic vegetation that can withstand waterlogged soil conditions has an advantage in establishing and occupying these areas [66]. Floods can also open up new areas for the recolonization of pioneer species. At the same time, different distances from the watercourse represent a combination of topography, flow rates, and soil properties, which together influence vegetation development processes and seed formation, distribution, dispersal, and deposition, leading to different responses of plant community structure to flooding [67, 68]. Merritt et al., (2010) achieved good results in predicting riparian occurrence and development by establishing the relationship between river flow and plant community response within different riparian zones. However, this method has limitations and has limited application [69]. Su et al., (2020) compared the characteristics of natural and unnatural floodplain plant communities in the Three Gorges reservoir and found that unnatural floodplain plants exhibited stronger ecological resilience [70]. Jian et al. (2018) systematically observed the trends of plant species in the riparian zone of Three Gorger reservoir from 2008-2015 and showed that the plant species composition was influenced by the new hydrological environment, identifying *Bermudagrass* or its community assemblages as the most suitable species for survival [71].

# 1.5.2. Soil physicochemical properties

The riparian buffer strips have typical interfacial effects and are a 'hot spot and moment' for various nutrients and biogeochemical cycles [72, 73]. Unique cycling characteristics in terms of soil water content, redox conditions, soil texture, nutrient availability, and microbial community at both vertical and horizontal scales due to significant habitat heterogeneity [72, 74]. As an important part of the riparian buffer zone, the soil is the central medium for all biological activities, and it provides material support for plants and regulates the effectiveness of nutrients and water [237]. In turn, vegetation cover, roots, litter, organic matter, and associated microbial communities affect soil porosity and other properties that influence soil moisture, infiltration, groundwater storage, and flow rates [75, 76]. Among them, soil physical structure such as soil water content, bulk density, which are considered as one of the essential indicators of

soil quality and soil productivity, and its chemical properties such as pH, organic matter, total nitrogen, rapidly available nitrogen, total phosphorus, rapidly available phosphorus, total potassium, rapidly available potassium, etc., which directly affect soil fertility and then, through influencing the growth of surface vegetation, ultimately affect the function of ecosystems of the water-land interface zone [77].

The function of riparian ecosystems is closely related to soil organic matter (SOM) turnover which is a complex and intertwined series of biological processes that cycle biological residues (e.g. plant litters, dead organisms, etc.) into inorganic molecules. The export of organic matter from the riparian zone to the river affects ecosystem metabolism and the rate of most biologically mediated reactions that regulate the fate of pollutants such as N, P, Hg, and pesticides in the riparian system [73]. With the water level fluctuation, the organic matter content in the riparian zone shows spatial heterogeneity. On the one hand, the organic matter in the river accumulates in the riparian ecotone with the water flow, and on the other hand, it will be carried away with the fall of water level, which is mainly related to the soil texture of the riparian zone [78]. Also, soil organic carbon is an important indicator for monitoring environmental changes in the riparian zone, Hale et al., (2014) found low variability in soil organic matter, total nitrogen and bulk density by monitoring soil physicochemical properties in different reaches of the watershed and concluded that these indicators have the potential for application to riparian zone monitoring [11].

Soil nitrogen and phosphorus cycling is a hot topic of widespread interest in riparian buffer strips [79], because these two elements have a direct relationship with water quality, and are also closely related to environmental problems such as soil acidification, eutrophication, and greenhouse gas emission. Fertilizers, pesticides, and insecticides applied by agricultural production from the uplands are considered to be the main source of nitrogen and phosphorus pollution, with excess loads entering rivers along surface waters, posing a potential threat to the water ecosystem [47, 80]. Riparian buffer strips are considered an effective way to remove nitrogen and phosphorus pollution, and numerous studies have shown that vegetation buffer strips of appropriate width can remove most agriculture pollution for uplands [5, 12, 50]. Long-term nutrient retention by vegetation buffer strips often results in N depletion and P enrichment, mainly because N can be temporarily conserved through vegetation uptake and denitrified into the atmosphere with the form of N<sub>2</sub>O and N<sub>2</sub>, which phosphorus does not have a long-term loss pathway and is more often adsorbed in the soil [81]. Nitrogen removal in riparian buffer strips is a complex process, with nitrate being highly mobile in water. Other forms of nitrogen, such as ammonium  $(NH_4^+)$ , are less susceptible to leaching because they are mostly bound to soil particles and resist the movement of water [82]. N leaching depends on the water-holding capacity of the soil and is more easily leaching in sandy soils with high permeability [83]. Microbial transformation of nitrogen includes assimilative uptake, reduction of assimilated nitrate to ammonium, reduction of dissimilated nitrate to  $NH_4^+$ (DNRA) or N gases such as dinitrogen (N<sub>2</sub>), nitric oxide, and through denitrification to nitrous oxide gas [8]. The pathway of phosphorus fixation and mobilization by vegetation buffer strips is relatively homogeneous, except for some soluble phosphorus active components that can be absorbed by vegetation, most of which are deposited within the buffers. Thus, vegetation buffer strips may be a source or sink of phosphorus pools. In ecosystems with low phosphorus content, vegetation buffer strips can be sink by trapping

phosphorus in surface runoff. While in phosphorus-saturated ecosystems, vegetation buffer strips may be a source, and regular harvesting of plants in the buffer strips to improve nutrient uptake by vegetation is necessary [84, 85].

#### **1.5.3.** Soil aggregates

Soil aggregate is the basic unit of soil structure and directly affects microbial community [86, 87], soil hydraulic properties [45], soil fertility status [88-89], soil erosion degree [90, 91], and so on. In addition, soil aggregate stability is one of the most important physical properties of soils, affecting water movement and storage, soil aeration, soil erosion, carbon cycle, biological activity, and crop growth [92]. Soil aggregate stability (SAS) is often used as an effective indicator to evaluate the fragmentation resistance of soil particles and the Mean Weight Diameter (MWD) is used to characterize the stability of soil aggregates; in general, the larger the MWD, the better the stability [91, 93]. At the same time, soil aggregates can increase soil organic carbon storage by reducing organic matter loss due to microbial mineralization and erosion [94]. Soil consists of large aggregates (>0.25mm) and microaggregate aggregate aggregation [95]. Soil physically protects organic carbon and promotes its stability by attaching particles of different sizes with organic and inorganic bonds [96].

The storage of organic carbon by soil aggregates is achieved through the mechanism of attachment of soil particle size to organic matter, and the relationship between different soil organic carbon forms and particle size has been widely revealed [95, 97]. A portion of free carbon is tightly bound to soil particles (<53µm) and mineral

elements by charge forces to form an inert carbon pool, which is not easily destroyed and can be preserved even for hundreds of years [98]. And a portion of organic carbon will be bonded with large particle size (>53µm) through colloids such as mycelium or extracellular secretions to form an active carbon pool, which is susceptible to turnover due to changes in environmental perturbations [99]. Hereby, Lavallee et al., (2020) advocated that ecologists should simplify the morphology of soil organic carbon into mineral-associated organic matter (MAOM) and particulate matter (POM) [100].

The stability of soil aggregates in riparian buffer strips is the result of combination of intrinsic and external factors [101]. External factors such as dry-wet moisture conditions and human disturbances decomposition of large aggregates through processes such as disintegration and fragmentation, while surface runoff carries fine particles towards rivers and changes soil structure, which in turn affects soil texture [234]. For example, Albalasmeh & Ghezzehei, (2014) and R. Ma et al., (2015) found that the overall stability of soil aggregates decreases with the increase in the number of dry-wet cycles [235, 236]. Soil physicochemical properties and binder materials also modulate the stability of soil aggregates through intrinsic effects, changing the contribution of binder material in stabilizing soil aggregates [101], and also affecting the carbon stability of soil aggregates of different particle sizes, accelerating the process of soil carbon cycle, which in turn affects the material turnover and energy flow in the riparian buffer strips [93]. For example, the dry-wet cycles will promote the exposure of clay-associated carbon to more biological interfaces for microbial use, reducing soil carbon stock change [87].

## 1.5.4. Microorganisms

Microbial communities play a key role in ecosystems and influence a vast array of important ecosystem processes, including biogeochemical cycling, organic matter decomposition, nutrient acquisition, pollutant purification, and soil structure maintenance [102]. Microorganisms are the main drivers of energy transformation and nutrient biogeochemical cycling in riparian ecosystems and are also important indicators of riparian ecosystem health [103]. Soil microbial in riparian zones are frequently affected by flooding (i.e., hydrological regimes) [104], resulting in dry-wet cycles and profoundly affecting the composition and function of soil microbial. For example, soil microorganisms species in the riparian zone were dominated by Proteobacteria and *Chloroflexi* at the phyla level, with a relatively small proportion of rare species. This suggests that the unique habitat of riparian zone has a strong filtering effect on microorganisms [102, 105]. Water-level fluctuation increases the complexity of the environment at the water-land interface, increases the abundance and diversity of soil microorganisms in the riparian buffer strips, and leads to a reduction in community stability [106]. Dry-wet cycle moisture conditions in riparian buffer strips also cause dramatic changes in the functional community of N-related microorganisms, altering the coupling relationship between nitrification and denitrification. For example, fluctuating groundwater levels in the riparian buffer zone produce alternating anoxic and hypoxic conditions that alternately promote nitrification and denitrification, thereby improving N removal through closely coupling N cycling processes [107-109].

Soil microorganisms in riparian buffer strips are shaped by a combination of

external environment, nutrient cycling, and soil physicochemical processes, and selection based on these physiological adaptations changes taxa abundance and genetic variation affecting community composition [110, 111]. Meanwhile, vegetation type is also an important factor influencing the structure of soil microbial communities in riparian buffer strips. The interactions between microorganisms are realized through indirect effects, on the one hand, microorganisms compete with plants for nutrients are regulate the nutrition and flora of above-ground vegetation; on the other hand, plants regulate soil structure through root extracellular enzymes and deposition, etc., and make directional selection on microbial community composition [112, 113]. Numerous studies have shown that soil microorganisms and vegetation communities in riparian zones are well adapted to each other and jointly regulate the ecological functions of riparian zones [114, 115]. In addition, soil microbial communities are sensitive to land-use type which not only characterize differences in nutrient input and output patterns of litters from above-ground vegetation type but also represent the effects of different soil management practices (disturbances) on microbial community structure. For example, natural forest ecosystems have higher soil microbial abundance the degraded frorest and agroecosystems [116]. The species richness and function of soil microbial communities in different successional land-use types have significant differences [117].

# **1.6.** Major factors affecting the environmental performance of riparian buffers

The marginal effects and wealth biodiversity of riparian buffer strips make them one of the most dynamic, diverse, and nature complexes on Earth. Meanwhile, riparian buffer strips are also fragile ecosystems that are highly vulnerable to hydrological, topographic, microclimatic, environmental, and anthropogenic disturbances [118]. Numerous lines of evidence from large scales such as buffer zone (>100m) suggest that hydraulic gradients due to distance from the watercourse are the most direct driver of riparian ecosystem heterogeneity [112, 119]. The vegetation, soil aggregate structure, physicochemical properties, and microbial community composition of the riparian zone will change accordingly depending on the distance from the watercourse [112]. For example, both soil organic carbon and nitrogen showed an increase with distance from the watercourse [120]. Riparian soil near rivers are characterized by coarse particles and low levels of organic matter [84]. Therefore, it is not known whether the buffer strips as a small region (<30m) exhibit similar characteristics on the distance scale.

At the local scale, the water level rhythm of storage is the main driver of soil and habitat change in the riparian buffer strips, while at the regional scale, disturbances from human activities and pollutant inputs indirectly affect the riparian buffer strips [121]. For example, soil bulk density tends to be higher in degraded riparian zones [122]. Soil bulk density may decrease as the effects of livestock trampling and heavy vehicles are eliminated after restoration, and vegetation coverage increases, organic matter (e.g. leaves and fine branches) accumulation and bare ground reduction after livestock exclusion and replanting leading to increased soil carbon content and improved soil structural stability [92, 123]. Ye et al., (2019) analyzed the change pattern of soil physicochemical properties in the Three Gorges reservoir riparian zone and found that soil physicochemical properties above 167.5 m elevation were more susceptible to the influence of regional anthropogenic disturbance [121].

In conclusion, the complex ecological environment of riparian buffer strips is the result of various environmental factors co-shaping including temperature, precipitation, topography, soil parent material, land management practices, and other factors. The relationship among these various factors is intricate and complex, and may even overlap in function. Wang et al., (2012) found that land-use type alone can explain about 30% of the variation in soil organic matter in reservoir's buffer strips through a comprehensive analysis of the effects of various environmental factors, while the inclusion of environmental factors explained only about 1% of the variation, suggesting that land-use type itself may also be the result of the combined effect of various environmental factors to the environmental performance of riparian buffer strips will help us to understand the complex soil-microbial-vegetation-environment relationships in riparian buffers and provide insights for riparian zone management and restoration.

# 1.7. Status of Chushandian Reservoir

The Huai River Basin is located in the eastern part of China between the Yangtze River and the Yellow River. The length of the mainstream is 1050 km, and the basin area is  $27 \times 10^4$  km<sup>2</sup>. Chushandian Reservoir is the only large-type reservoir built in the upper reaches of the main stream of the Huai River. The reservoir is a large-scale water conservancy project constructed mainly for flood control, irrigation, water supply, and power generation. After decades of construction, the Chushandian reservoir was finally officially launched for water storage in May 2019 and reached baseline (normal) water level for the first time in October 2020. The reservoir's location is mainly based on the

original channel of the Huai River, and the surrounding area of the original channel has been widened, are nearly 40,000 residents have been relocated. The land-use types around the reservoir in the early period were mainly cropland and commercial woodland (agroforest ecosystem with Tea and Chestnut interplanting). During the construction of the reservoir, some of the cropland was abandoned and gradually evolved into grassland (natural recovery), and some of the cropland was not abandoned for cultivation until the water level arised. Therefore, after reservoir impoundment, a mosaic landscape pattern dominated by abandoned cropland, grassland, and commercial woodland was formed around the shoreline. Since the different land-use types were previously operated by scattered farmers, the landscape was fragmented and the width of independent land-use type was usually between 25-30m, which was the basis for the next step of the reservoir's buffer strip construction.

## **CHAPTER 2**

#### METHODOLOGY

#### 2.1. Study area

We carried out our experiment along the reservoir's buffer strips of the Chushandian Reservoir (N32 22'-32 32', E113 89'-113 96') in Xinyang City, Henan Province, China. Xinyang is a transitional region from a subtropical climate zone to a warm temperate zone located in the southern part of Henan Province. The city is located at the dividing line of the Qinling and Huai Rivers. The terrain is high in the south and low in the north, with an altitude of 75-300 m. The annual average temperature is generally between 15.3-15.8 °C, and the annual average rainfall is 993-1,294 mm. The air is moist and the relative humidity ranges from 74-78% annually (China Meteorological Data Sharing Service System, http://data.cma.gov.cn). There are many rivers in this region, which belong to the Yangtze River catchment and the Huai River catchment, and the Huai River basin accounts for 98.2% of Xinyang City's area. Due to the influence of Xinyang's particular geography and climate, the spatial and temporal distribution of rainfall is uneven, and precipitation varies greatly within and between years. Xinyang's precipitation mainly comes in June to August, and the difference in precipitation between wet and dry years can be a factor of up to 3. The main forest tree species in this area are Pinus massoniana Lamb, Cunninghamia lanceolata (Lamb.) Hook., Pistacia chinensis Bunge, and Quercus dentata Thunb., among many others. These tree species are distributed in pure forests or mosaics and form the main surrounding forest community [124].

# 2.2. Field investigation

We conducted our soil survey on December 11, 2020, which was two months after the first controlled flood of the reservoir area reached its predetermined normal storage level. The local soil texture type was mainly *ACRISOLS* soil (based on the FAO World Reference Base) with heavy soil viscosity and weak acidity.

Along the western bank line of the reservoir (without dike protection) we selected three land-use types (abandoned cropland, grassland, and woodland), and three plots of similar conditions were selected as replicates for each land-use types (9 in total) (Figure 2.1). Up to the time of sampling, the abandoned cropland had naturally recovered over main vegetation was Veronica didyma Tenore, several months. The Convza canadensis (L.) Crong., and Alternanthera philoxeroides (Mart.) Griseb. The grassland was previously abandoned cropland (no agricultural production during the initial stage of reservoir construction due to the relocation of residents). At the time of sampling, the sample site had undergone 4-5 years of natural recovery, and the main vegetation was Imperata cylindrica (L.) Beauv., Xanthium sibiricum Patrin ex Widder, and Conyza canadensis (L.) Cronq. The woodland adopts typical agroforest ecosystems with an excellent local history, the main species of tree was chestnut (Castanea mollissima BL.), and the understory was the tea variety Xinyang Maojian (Camellia sinensis (L.) O. Ktze.), and the main vegetation was Imperata cylindrica (L.) Beauv. and Carex Linn., *Phyllostachys glauca* McClure (Table 2.1).



Figure 2.1 Three land-use types around the Chushandian reservoir's buffer strips.

Table 2.1

Land-use types	Previous	Recovery time	Slope	Dominate Vegetation
Abandoned cropland	cropland	6-7 months	0	Veronica didyma Tenore; Conyza canadensis (L.) Cronq.; Alternanthera philoxeroides (Mart.) Griseb.
grassland	cropland	4-5 years	0	Imperata cylindrica (L.) Beauv.; Xanthium sibiricum Patrin ex Widder;Conyza canadensis (L.) Cronq.
woodland	woodland	more than 10 years	13	Castanea mollissima BL.; Camellia sinensis (L.) O. Ktze.

#### Basic state of different land-use types.

The sampling strip distance was set according to the actual plot size of different land-use types along the watercourse of the reservoir. As the different historical land-use types were previously operated by a patchwork of farmers, the landscape was fragmented, and the width of independent land-use types ranged from 25 to 30 m. To eliminate the influence of boundary effects of adjacent sample plots, we set up sampling strips of 0 m, 2 m, and 20 m within different land-use types along the bank line according to the distance from the watercourse (Figure 2.2). For the convenience of expression, we wrote land-use types and distance scales together as woodland: W0, W2, W20; grassland: G0, G2, G20; abandoned cropland: C0, C2, C20. We collected 27 soil samples in total.

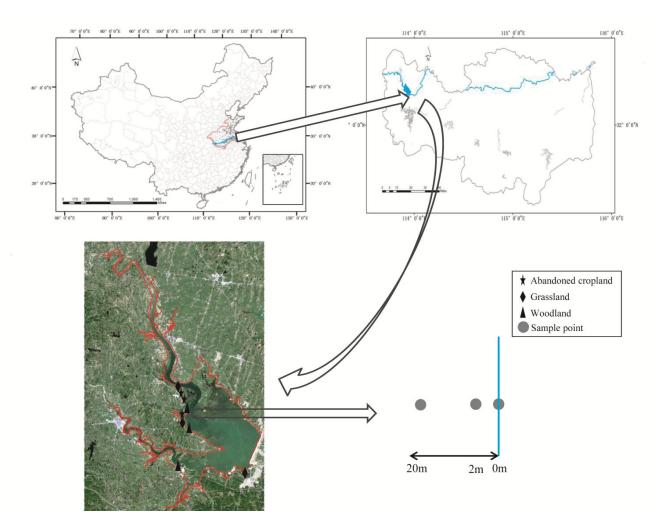


Figure 2.2 Location of the study area and research design.

# 2.3. Soil sampling

Taking the boundary between the watercourse and land as a sampling point of 0 m, we took our samples using a *Petersen* grab, and three adjacent soil samples were selected for each location and mixed into one. At 2m and 20m, we collected 5 soil cores intactly near the sampling point, mixed them into a homogeneous composite sample, and kept an additional soil core intact that was immediately brought back to the laboratory for measurement of BD (soil bulk density) and SWC (soil water content). We did not measure the physical properties of the soil at 0m because the soil at this location is at the

land-water interface and it was too difficult to collect a complete soil core intact. All composite samples were divided into two sections. One was kept in a self-sealed bag and placed in a refrigerator at 4 °C for the determination of  $NH_4$ -N,  $NO_3$ -N, DOC, MBC, and MBN content. And the other one was stored in a self-sealed bag and brought back to the laboratory for air drying. Stones and roots were picked out with forceps, and separated into two portions using a 2 mm sieve. One portion was used to determine the soil particle structure, and the other was screened through a 0.25 mm sieve and refrigerated at 4 °C for pH, TC (total carbon), TN (total nitrogen), and TP (total phosphorus) determination. All tests were conducted within two weeks after sampling.

#### 2.4. Vegetation survey

Strips transect (parallel to the stream direction) were set up at 2m and 20m in each sampling plot, and three 1m × 1m sub-sample squares were set up in each strips transect, with 10m interval between squares. Vegetation survey was conducted in each square, and the species, number of each species, and coverage of all plants were recorded. Then, Refer to the <Flora in China>, <Flora in Henan Province>, <Atlas of Higher Plants in China>, <Altas of Rare and Endangered Plants in China>, and other works on botany for identification.

#### 2.5. Laboratory analysis

#### 2.5.1. Soil physicochemical properties

We determined the level of TC and TN in our soil samples by elemental analyzer

(Vario MAX C/N, Elementar Analysensysteme GmbH, Hanau, Germany), and soil pH was measured in distilled water mixed 2.5:1 (by volume) with dry soil by a Delta 320 pH meter (Mettler-Toledo Instruments (Shanghai) Ltd., China). We determined soil TP content by using the molybdenum-blue colorimetry method after digesting the samples with perchloric acid. Both NH<sub>4</sub>-N and NO<sub>3</sub>-N were extracted using 1 mol/L KCl, using a soil: KCl ratio of 1:10 and were both measured using an Automatic Discontinuous Chemical Analyzer (Smarthem200, Alliance Company, French). We extracted DOC with K<sub>2</sub>SO<sub>4</sub> and determined its level by dichromate digestion. MBC and MBN were measured by the fumigation extraction method [125, 126], and we measured BD (bulk density) using the oven-drying volumetric ring method after samples were oven-dried at 105 °C for 24 hours to a constant mass. We then calculated BD as the ratio of oven-dried undisturbed core weight to the cutting ring volume. SWC was determined by oven-drying the samples at 105 °C for 24 hours, and the water content was expressed as a percentage of the dry weight.

#### 2.5.2. Particle-size fraction and associated carbon separation

In this study, we physically grouped soil particles by size with reference to the criteria of the soil texture classification system: > 53  $\mu$ m (sand particle), 2-53  $\mu$ m (silt particle), and < 2  $\mu$ m (clay particle). We isolated particles from composite samples using the method proposed by Tiessen (1982) and Wu et al. (2004) [127, 128]. First, we weighed 20 g of soil that passed through a 2 mm sieve into a 250 mL beaker, and then we placed the soil in a sonifier to discretize it fully for 30 min. Next, we poured the suspension into a 53  $\mu$ m sieve and washed it with about 450 mL of distilled water until

the washout became clear. The material left on the top of the sieve was sand particles (>53 µm) and some plant residue. After this, we placed approximately 300 to 350 mL of the eluate into a 450 mL centrifuge tube and spun it at 760 rpm for 4 min using a Mandal RC5C centrifuge. We then poured the suspension into a collector and added 100 mL of distilled water. We then shook it and further centrifuged it at 550 rpm for 2 min, decanted the suspension, and combined it with the previous suspension. The latter process was performed at least 4 or more times until complete separation of the clay particle (< 2 µm) from the silt particle (2 to 53 µm) is achieved. We placed all collected particles in an oven at 50°C for 72 hours and then weighed them. Finally, we determined the carbon and nitrogen content of each particle using an elemental analyzer (Vario MAX C/N, Elementar Analysensysteme GmbH, Hanau, Germany).

#### 2.6. Microbial diversity analysis

#### 2.6.1. DNA extraction and PCR amplification

Microbial DNA was extracted from soil samples using the E.Z.N.A.® soil DNA Kit (Omega Bio-tek, Norcross, GA, U.S.) according to manufacturer's protocols. The final DNA concentration and purification were determined by NanoDrop 2000 UV-vis spectrophotometer (Thermo Scientific, Wilmington, USA), and DNA quality was checked by 1% agarose gel electrophoresis. The V3-V4 hypervariable regions of the bacteria 16S rRNA amplified primers gene with 338F were (5'-ACTCCTACGGGAGGCAGCAG-3') and 806R (5'-GGACTACHVGGGTWT CTAAT-3') by thermocycler PCR system (GeneAmp 9700, ABI, USA). The PCR

reactions were conducted using the following program: 3 min of denaturation at 95 °C, 27 cycles of 30 s at 95 °C, 30s for annealing at 55 °C, and 45s for elongation at 72 °C, and a final extension at 72 °C for 10 min. PCR reactions were performed in triplicate 20  $\mu$ L mixture containing 4  $\mu$ L of 5 × FastPfu Buffer, 2  $\mu$ L of 2.5 mM dNTPs, 0.8  $\mu$ L of each primer (5  $\mu$ M), 0.4  $\mu$ L of FastPfu Polymerase and 10 ng of template DNA. The resulted PCR products were extracted from a 2% agarose gel and further purified using the AxyPrep DNA Gel Extraction Kit (Axygen Biosciences, Union City, CA, USA) and quantified using QuantiFluor<sup>TM</sup>-ST (Promega, USA) according to the manufacturer's protocol.

# 2.6.2. Illumina MiSeq sequencing

Purified amplicons were pooled in equimolar and paired-end sequenced  $(2 \times 300)$  on an Illumina MiSeq platform (Illumina, San Diego,USA) according to the standard protocols by Majorbio Bio-Pharm Technology Co. Ltd. (Shanghai, China). Raw sequences were processed using QIIME2 pipeline, quality control, and denoised with DADA2, obtained Absolute Sequence Variants (ASVs) which desponded on 99% similarity. The taxonomy classification was carried out using the Silva database (http://www.arb-silva.de/) as a reference. We used the metagenome prediction tools PICRUST2 to predict the function profiles of microbial communities based on their taxonomic composition. The ASVs table was normalized by count, and then the predicted functional gene community was generated using the Kyoto Encyclopedia of Genes and Genomes (KEGG) database. The predicted functional metagenomes were then classified at the KEGG 3 level.

### 2.6.3. Quantification of bacterial community assembly

The mean nearest taxon distance (MNTD) was a terminal metric of phylogenetic diversity that could quantify the phylogenetic turnover based on the phylogenetic distance among the closest relatives [106]. The nearest taxon index (NTI) was evaluated by the standard deviation that represented the MNTD value from the mean of the null distribution (999 randomizations) to quantify the degree of nonrandom processes in the assembly of the microbial community. If a mean NTI taken from a given group is significantly different than zero, that NTI indicates phylogenetic clustering (i.e., the coexisting taxa are more closely related than they are expected to be by chance (NTI > 0) or overdispersion (NTI < 0) (i.e., coexisting taxa are more distantly related than they are expected to be by chance).

Null model analysis was used to detect the relative importance of stochastic and deterministic processes in mediating microbial community succession to quantify community assembly [129]. In brief, we first calculated the beta nearest taxon index ( $\beta$ NTI) to measure the relative contribution of different assembly processes. A significant deviation ( $|\beta$ NTI| > 2) indicated the dominance of selection processes (deterministic processes) during the succession of the community. Among these,  $\beta$ NTI > 2 represented variable selection while  $\beta$ NTI < -2 represented homogeneous selection. On the contrary,  $|\beta$ NTI| <2 meant that the stochastic process was important in community assembly [129]. Subsequently, Bray–Curtis-based Raup–Crick (RCbray) was used to further partition the stochastic processes ( $|\beta$ NTI| < 2). The relative influence of homogenizing dispersal was

quantified as the fraction of the ecological process with  $|\beta NTI| < 2$  and RCbray < -0.95. Dispersal limitation was quantified as the fraction of process with  $|\beta NTI| < 2$  and RCbray > 0.95.  $|\beta NTI| < 2$  and |RCbray| < 0.95 indicated "undominated," suggesting that community variations were driven by weak selection, weak dispersal, diversification, and/or drift [129-130].

# 2.7. Yeild and economic productivity investigate

Investigated the yeilds and economic incomes of chestnut and tea from the farmers who adopted agroforest ecosystems in the reservoir's buffer strips by way of field inquiry, The main contents include planting time, planting cost, harvest yields, and economic incomes. Meanwhile, investigated the yeilds and economic incomes of the farmers who only cultivated chestnut or tea in nearby highlands, respectively, and compared the difference of yeilds and economic incomes between adopted agroforest ecosystems and traditional monoculture.

# **CHAPTER 3**

# THE VEGETATION BIODIVERSITY FOR ADOPTED AGROFOREST ECOSYSTEMS IN RESERVOIR'S BUFFER STRIPS

#### 3.1. Statistical analysis

Calculation of vegetation biodiversity indices (*Important Value, Relative density, Relative frequency, Relative coverage, Simpson index, Richness index, Shannon index, Pielou index*) for reservoir's buffer strips with three land-use types based on survey results. The vegetation biodiversity indices were calculated using the following formula [117].

Important value = (Relative density + Relative frequency + Relative coverage) / 3

Where *Relative density* = (Number of individual plant species / Number of species in all plots); *Relative frequency* = Frequency of individual plant species present / The total frequency of all plant species; *Relative coverage* = The fraction coverage of an individual plant species in plot / Total coverage of all individual plant species in plots.

Richness Diversity Index (S):

S = Pa

Simpson Diversity Index (Simpson):

Simpson =  $1 - \sum (P_i)^2$ 

Shannon Diversity Index (Shannon):

 $Shannon = -\sum P_i \ln P_i$ 

Pielou Diversity Index (*J*):

 $J = Shannon / \ln S$ 

Where  $P_a$  is the number of the species in the sample.  $P_i = N_i/N$ ,  $N_i$  is the number of individuals of the *i*th species in the sample, N is the total number of individuals in the plots.

We performed an analysis of variance (ANOVA), and post-hoc multiple comparisons (Tukey) tests at the probability levels (p) of 0.05 to determine differences between the soil vegetation biodiversity in reservoir buffer strips with different land-use types.

#### **3.2. Results**

3.2.1 Vegetation species composition in reservoir's buffer strips with three land-use types

Through investigation, vegetation species in Chushandian reservoir's buffer strips were abundant, and 58 species, 54 genera, and 29 families of vascular plants were identified. Among them, *Compositae* 10 sepcies, 8 genera; *Rosaceae* 5 species, 4 genera; *Gramineae* 5 species, 5 genera; *Labiatae* 4 species, 4 genera; *Liliaceae* 3 species, 3 genera; *Leguminosae* 3 species, 3 genera; *Scrophulariaceae* 3 species, 3 genera; *Caryophyllaceae Juss* 2 sepcies, 2 genera; *Mbelliferae* 2 sepcies, 2 genera; *Euphorbiaceae* 2 species, 1 genera; *Pteridaceae*, *Callitrichaceae*, *Cyperacea*, *Geraniaceae*, *Lycopodiaceae*, *Fagaceae*, *Menispermaceae*, *Ericaceae*, *Urticaceae*, *Brassicaceae*, *Convolvulaceae Juss*, *Moraceae*, *Primulaceae*, *Juncaceae* all were 1 species, 1 genera. *Compositae* was the most abundant species, followed by *Rosaceae* and

*Gramineae*, which accounted for 17.24%, 8.62% and 8.62% of the total species, respectively, and were the main dominant species in the reservoir's buffer strips.

There were obvious differences in vegetation species in the reservoir's buffer strips for three land-use type, among which the species in woodland was the most abundant land use types as high as 27, probably due to the low human interference and good growing environment, which means that the adopted agroforest ecosystems in the reservoir buffer strips can increase the vegetation biodiversity. While, There were 22 species in abandoned cropland and 20 species in grassland, respectively. The results indicated that the low elevation for abandoned cropland and grassland may be flooding for a long time, and the number of the vegetation species adapted to the environment is relatively small.

It can be seen from Appendix B that among the 20 species in the grassland, the *Erigeron annuus, Imperata cylindrica,* and *Veronica persica* are the coexisting species of three different land types. The species of *Carex breviculmis, Cerastium glomeratum Thuill., and Conyza canadensis* are co-existing in grassland and abandoned cropland. With the living environment change, the number of species living in different land use also changes accordingly. Compared with the grassland, the abandoned cropland have more 16 species, such as *Mazus pumilus, Cirsium setosum, Digitaria sanguinalis, Lagopsis supina*, etc., among which 11 species, such as *Lamium amplexicaule L., Alternanthera philoxeroides* and *Artemisia princeps*, were only presented in abandoned cropland. There were Twenty-two vegetation species, such as *Kalimeris indica, Rhododendron simsii Planch., Rosa chinensis Jacq.*, and *Phyllostachys glauca McClure*, are found only

in woodland.

**3.2.2.** Vegetation biodiversity in reservoir's buffer strips with three land-use types

The vegetation biodiversity (*Simpson*, *Shannon*, *Richness*, *Pielou index*) of three land-use types all showed grassland is lower than the other two land-use types though did not differ significantly among land-use types (Figure 3.1). The *Simpson*, *Shannon* and *Richness* index of C2 was significantly greater than C20, the other two land-use types showed the same trend in distance but were not significant. This indicates that the vegetation community of abandoned cropland is more susceptible to flooding, and the vegetation community population changes dramatically.

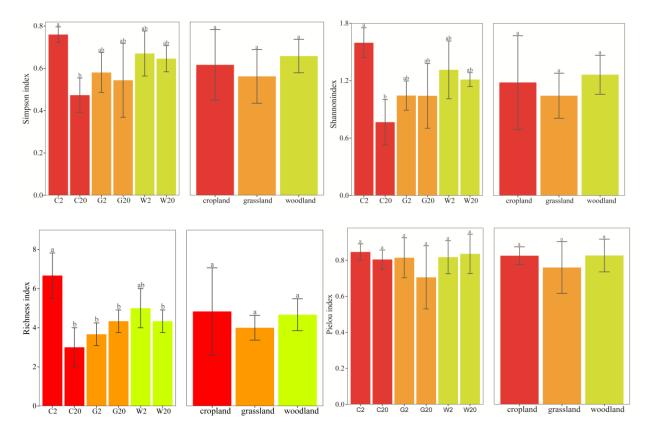


Figure 3.1 Variation of vegetation biodiversity indices (Simpson index, Shannon

index, Pielou index, Richness index).

# **3.3.1.** Vegetation species composition in reservoir's buffer strips with three land-use types

The original habitat conditions of the riparian buffer strips in Chushandian reservoir were completely destroyed after the dam construction. The water level fluctuation changed the growth environment of the vegetation composition and the community population also alternating under of the dry-wet cycles, and the native vegetation population deteriorated seriously, and the vegetation biodiversity also decreased [131]. In this study, we investigated three different land-use types and found 58 species of vascular plants in 29 families, and 54 genera, and the phenomenon of single species and genera was obvious. Among these three land-use types, the species of Erigeron annuus, Imperata cylindrical, and Veronica persica Poir. were coexisting, and the community composition was simplified at 2m of the three land-use types. The reason why annual plants became the dominant species may be that they were affected by flooding stress, and the structure and function of vegetation adapted to this environment were able to grow [65]. Therefore, at 2 m for different land-use types where the flooding time is longer, annual herbs are mainly present, while at 20 m away from the flooding area, the flooding time is shorter, which is more conducive to the growth of perennial plants. This may be due to the increasing distance, the weakening of the flooding stress of the vegetation in the reservoir buffer strips, the proportion of perennial plants increased significantly, and the species and population of trees and shrubs also began to increase. In addition, due to the proximity of 20m to the terrestrial environment above the

reservoir' s buffer strips, various terrestrial seeds from the upland also burst into the area through animals and wind media, which is also the reason for the increase of perennial plants. It can be foreseen that under the condition of seasonal water level fluctuation in Chushan Reservoir, the life forms of herbaceous vegetation (including annual and perennial plants) replace the life forms of grass and shrubs, which is an inevitable trend of the change of vegetation life composition in the reservoir's buffer strips of Chushandian Reservoir [132].

# **3.3.2.** Vegetation biodiversity in reservoir's buffer strips with three land-use types

The alternations of seasonal change of water level eventually evolved into the riparian zone, and the vegetation in riparian buffer strips formed secondary bare land growth and reproduction during the seasonal flooding time, which led to the change of the growth mode and population of the vegetation community with the change of habitats, and the distance from the watercourse was an important factor affecting the diversity of the vegetation community [69].

Through investigation, it was found that *Compositaceae*, *Rosiaceae*, and *Gramineae* were the dominant species in three land-use types. Annual plants have become the dominant species because the growth environment is harsh under the seasonal alternating pattern of flooding and drought. Compared with annual plants, perennial plants have a longer physiological cycle and need a longer period of water retreat and storage to complete their life history [119]. We also found that the vegetation biodiversity in 2 m was higher than 20 m, especially in abandoned cropland, which indicated that the

vegetation species composition and population change more dramatically in the near water area, this may correlate with the frequency of water flooding. Numbers study have demonstrated that the water level fluctuation will create a hotspot of biodiversity for plants and animals [106, 134].

Generally speaking, the greater the degree of plant flooding stress, the lower the plant diversity. The Richness, Simpson, and Shannon index and dominance of vegetation in the riparian buffer strips of Chushandian Reservoir show different trends, and the diversity index of woodland is the highest, which may be due to the fact that the ground is less disturbed by human beings and suitable for plant growth. There are more resources available than abandoned cropland and grassland, among which shrubs and small trees also appear relatively. Among them, the dominance degree of the annual Pendergrass and white grass communities is more obvious. Due to the long time of flooding and harsh growth environment in the 2m riparian buffer strips, only a few water-flooding resistant species (such as the *Cerastium Glomeratum Thuill.*) can form stable communities.

### **3.4.** Conclusion

In this study, different land-use types have a greatly influence on vegetation biodiversity in the riparian buffer strips of Chushandian reservoir. The main plants in the riparian buffer strips are *Compositae*, *Roseaceae*, and *Gramineae*, and the phenomenon of single genus and species is obvious, and the community composition is simplified. The vegetation at 2m distance from the watercourse was mainly annual plants. At 20m, plant life gradually diversified, perennial plants generally increased, and trees and shrubs also began to increase. Among them, are the species of *Erigeron annuus*, *Imperata cylindrical*, and Veronica persica Poir. were coexisting in these three land-use types.

As for agroforest ecosystems, the distance to the watercourse obviously was an ignored factor when considering management. While compare to abandoned cropland, the agroforest ecosystems seem to have some capacity to maintain the stability of vegetation community structure. We also found that agroforest ecosystems can support more plant species, even including some shrubs, which indicated that adopted agroforest ecosystems in the reservoir buffer strips can promote more plant productivity.

### **CHAPTER 4**

# THE SOIL PROPERTIES FOR ADOPTED AGROFOREST ECOSYSTEMS IN RESERVOIR'S BUFFER STRIPS

#### 4.1. Statistical analysis

To reveal more comprehensively the relationship between soil physicochemical properties, all soil parameters obtained were classified into soil properties (includeing pH, TC, TN, TP, NH<sub>4</sub>-N, NO<sub>3</sub>-N), soil texture (including sand content, silt content, clay content), soil stoichiometric ratio (include C/N, MBC/MBN), and microbial activity (include MBC, MBN, DOC) before performing data analysis. We performed an analysis of variance (ANOVA), and post-hoc multiple comparisons (Tukey) tests at the probability levels (p) of 0.05 to determine differences between the soil physicochemical properties in the reservoir buffer strips with different land-use types. Two-way ANOVA was performed to determine the interaction between land use types and the distance from the watercourse. Pearson's correlation coefficient was used to analyze the relationship between soil properties at significance levels of 0.05 and 0.01. The mantel test was used to calculate the relationship between soil texture, soil stoichiomotric ratio, microbial activity, vegetation biodiversity indices, and soil property parameters. In addition, we used principal component analysis (PCA) to explore the relationship between different soil parameters and vegetation biodiversity indices in the reservoir's buffer strips with three land-use types and distances from the watercourse. Redundancy analysis (RDA) was used to explore the main factors affect soil properties. Specifically, the variables with significant effects on soil properties were first identified using a combination of forward

and backward selection, and then the explanatory rate, as well as significance, was calculated for the screened variables using the *rdacca.hp* package. Structural equation models (SEM) between land-use types, distance from the watercourse, soil texture, soil properties, soil stoichiometric ratio, soil microbial activity, and vegetation biodiversity indices were constructed using a partial least squares path model (PLS-PM). Barplots and residuals were constructed to check the common assumptions of homogeneity of variances, normality for all models, and lack of collinearity for multiple regression models, and variables were log10 transformed if necessary. All analysis was computed using R version 4.0.3. The main related packages used are *ggplot2*, *vegan*, *tidyverse*, *plsm* etc.

#### 4.2. Results

# 4.2.1. Soil physicochemical properties in reservoir's buffer strips with three land-use types

The reservoir's buffer strips with three land-use types were weakly acidic, with pH values varied in the range of 5-6.5; The soil TC and TN at W2, which were 26.08 and 2.13 g/kg, respectively were significantly higher than those at other sites, and the soil TC and TN in abandoned cropland and grassland, when W0 was not considered, showed a gradual decrease (although not significant) with increasing distance from the watercourse. Soil TC and TN were significantly higher in woodland while TP was significantly lower than the other two land-use types (Figure 4.1); The content of NH<sub>4</sub>-N at W20, which was 45.31 mg/kg, was lower than that at other sites, and the content of NO<sub>3</sub>-N at W2 and

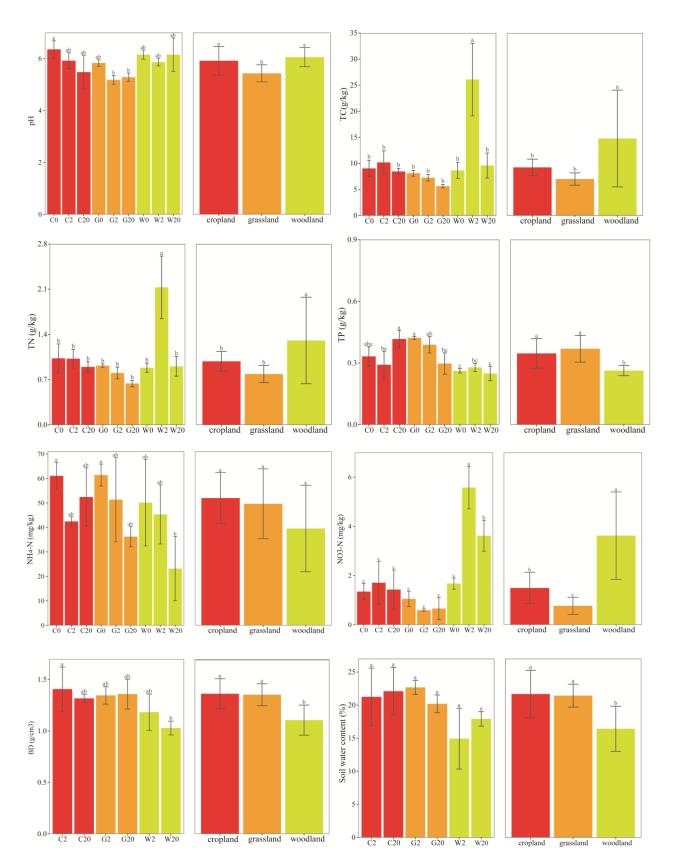


Figure 4. 1 Variation of soil properties (pH, TC, TN, TP, NH<sub>4</sub>-N, NO<sub>3</sub>-N, BD, SWC).

W20, which were 5.58, 3.62 mg/kg, respectively, was significantly higher than the other

sites (p<0.05), and the content of NO<sub>3</sub>-N varied significantly between three land-use types, showing woodland > abandoned cropland > grassland; Soil bulk density (BD) at W20, which was  $1.03 \text{ g/cm}^3$ , was lower than all other sites, and the BD and SWC of woodland were significantly lower than the other two land-use types (Figure 4.1).

The contents of microbial biomass carbon (MBC), microbial biomass nitrogen (MBN), and dissolved organic carbon (DOC) at W20, which were 418.89, 35.99, and 23.38 mg/kg, respectively, were significantly higher than those at the other sites (p<0.05), the content of MBC, MBN, DOC were significantly higher in woodland than grassland (Figure 4.2).

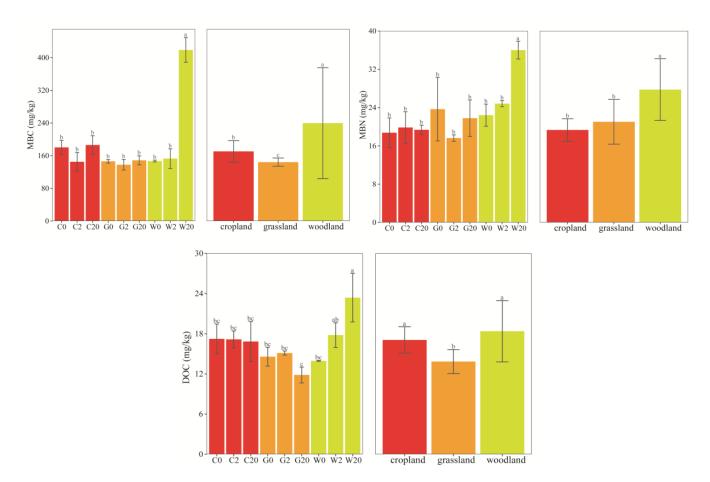


Figure 4.2 Variation of soil microbial activity (MBC, MBN, DOC).

At 51.11%, the sand content of woodland soil was higher than that of the other two land-use types. However, the silt content of woodland soil was significantly lower than that of the other two land-use types (p<0.05) at only 34.75% (Figure 4.3). The clay content did not differ significantly among the three land-use types. The soil C/N of woodland was 10.82, which was significantly higher than the other two land-use types. The soil MBC/MBN of the abandoned cropland was 8.96, which was significantly higher than that of grassland (Figure 4.4).

Table 4.1

Soil	Historical	Land-use types	Distance	from water	Types × Distance	
properties	F value	P value	F value	P value	F value	P value
pН	3.66	0.028 *	2.60	0.121	-	-
ТС	3.47	0.034 *	4.47	0.046 *	-	-
TN	4.12	0.018 *	3.02	0.096	-	-
ТР	0.15	0.927	0.45	0.508	3.82	0.039 *
NH <sub>4</sub> -N	0.70	0.564	6.15	0.021 *	-	_
NO <sub>3</sub> -N	10.23	<0.001***	0.15	0.703	-	-
DOC	9.71	<0.001***	2.02	0.170	13.34	< 0.001***
MBC	39.34	<0.001***	114.66	<0.001***	92.64	< 0.001***
MBN	14.72	<0.001***	8.14	0.010 **	7.19	0.004 **
Sand	17.18	<0.001***	0.76	0.614	-	-
Silt	11.43	<0.001***	0.93	0.502	-	-
Clay	1.27	0.315	0.18	0.978	-	-
BD	7.26	0.007 **	1.54	0.235	-	-
SWC	5.41	0.018 *	0.09	0.764	-	-
C/N	5.77	0.005 **	0.09	0.762	-	-
MBC/MBN	2.80	0.086	9.44	0.006 **	7.46	0.004 **

### The correlation and interaction of soil physicochemical properties between historical

land-use types and distance from watercourse (k=6, n=3).

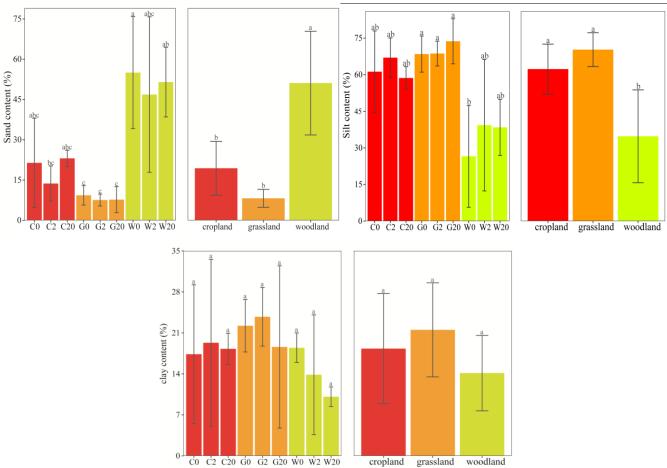


Figure 4.3 Variation of soil texture (Sand content, Silt content, Clay content).

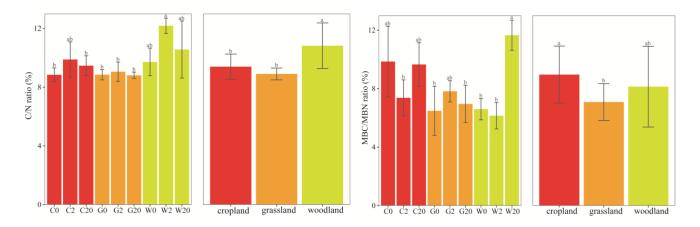


Figure 4.4 Variation of soil stoichiometric ratio (C/N, MBC/MBN).

According to the results of the two-way *ANOVA* (Table 4.1), we found that the physicochemical parameters of the reservoir's buffer strips with three land-use types were affected by land-use types except for TP, NH<sub>4</sub>-N, Clay content, and MBC/MBN. While,

MBC and MBN also received effects from both distance from the watercourse (F=114.66, p<0.001; F=8.14, p=0.01), and interaction between them (F=92.64, p<0.001; F=7.19, p=0.004). NH<sub>4</sub>-N and MBC/MBN were only affected by the distance from the watercourse (F=6.15, p=0.021; F=9.44, p=0.006).

# 4.2.2. The relationship between physicochemical properties and vegetation biodiversity in reservoir's buffer strips with three land-use types

Our analysis shows that TN was extremely positively correlated with TC (0.91, p<0.01), and both of these were positively correlated with NO<sub>3</sub>-N and pH. pH was extremely positively correlated with NO<sub>3</sub>-N and BD (p<0.01). TP was positively correlated with NO<sub>3</sub>-N and negative with SWC (p<0.05). NH<sub>4</sub>-N was positive correlated with BD and negative with SWC (p<0.05). Soil microbial activity was extremely correlated with NH<sub>4</sub>-N, soil stochiometric ratio was extremely correlated with NO<sub>3</sub>-N, and vegetation biodiversity indices were extremely correlated with BD and SWC (Mantel's p<0.01) (Figure 4.5).

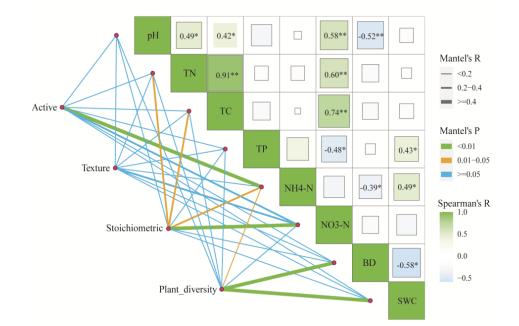


Figure 4.5 Pairwise comparisons of soil properties are shown, with a color gradient denoting Spearman's correlation coefficients.

It can be seen from the PCA results that different sites of the reservoir's buffer strips with three land-use types were well distinguished, with 0m site of three land-use types all clustered together and positively correlated with SWC and NH<sub>4</sub>-N. The 2m and 20m sites of woodland clustered together, and were positively correlated with TC, TN, sand content, soil microbial activity, and stoichiometric ratio. The 2m and 20m sites of grassland and abandoned cropland were not clearly distinguished on the first component axis, while were well separated on the second component axis. Grassland was positively correlated with Silt content, Clay content, and TP, and abandoned cropland was positively correlated with vegetation biodiversity indices (Figure 4.6).

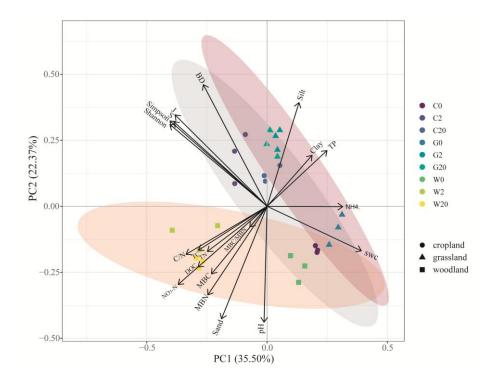


Figure 4.6 PCA (principal component analysis) biplot representing the relationship between the soil physicochemical parameters and vegetation biodiversity indices.

The results of the RDA analysis showed that soil microbial activity, soil texture, soil stoichiometric ratio, and vegetation biodiversity indices could explain 47.54% of the variation of soil properties, with 38.7% in the first axis. Simpson index, Distance from the watercourse, MBN, Silt content, and C/N had strong explanations for soil properties (Figure 4.7), with Simpson index, MBN, and Distance from the watercourse reaching significant levels (Figure 4.8).

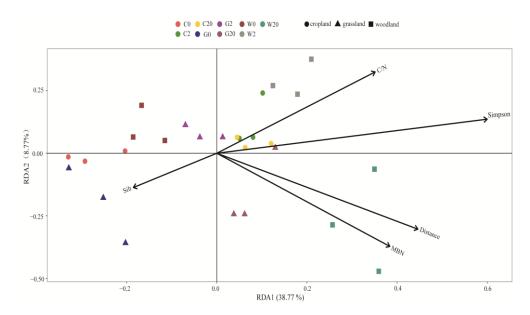


Figure 4.7 RDA (redundancy analysis) using soil microbial activity, soil texture, soil stoichiometric ratio and vegetation biodiversity indices to explain soil properties.

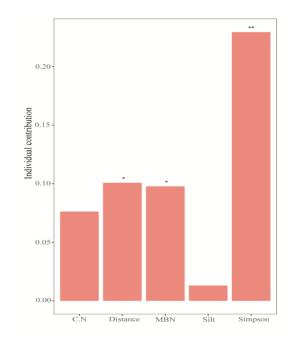


Figure 4.8 The explanatory ratio and significance of variables selected from RDA.

A more in-depth analysis using a PLS-PM showed direct and indirect effects of land-use types and distance from the watercourse on soil texture, soil properties, soil microbial activity, soil stoichiometric ratio, and vegetation biodiversity indices (Figure 4.9). Land-use types directly affected microbial activity, soil texture, and stoichiometric ratio, whereas land-use types affected soil properties indirectly through stoichiometric ratio. Distance from the watercourse directly affected microbial activity and vegetation biodiversity indices, whereas distance from the watercourse affected soil properties indirectly through vegetation biodiversity indices. The goodness of fit (GOF) was 0.560, and indicating that the model has relatively good predictive power.

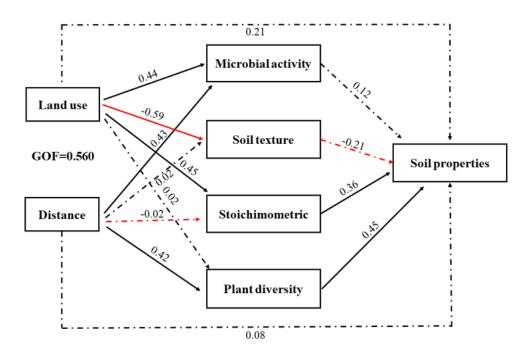


Figure 4.9 Causal inference diagram based on PLS-PM.

#### 4.3. Discussion

Our study focused on the differences between soil physicochemical properties and the relationship between soil physicochemical parameters and vegetation biodiversity indices at fine-scale in the reservoir's buffer strips with three land-use types, explored the variation patterns of soil physicochemical properties at fine distance scales, revealed the main factors affecting soil properties, and provided new insights for understanding the reservoir's buffer strips, especially in the context of weighing the contradiction between agricultural production and ecological benefits [1], and provided valuable theoretical references for adopted agroforest ecosystems in the reservoir's buffer strips. 4.3.1. Differences of soil physicochemical properties for three land-use types in reservoir's buffer strips

As the last barrier to prevent soil nutrient loss, buffer strips play an essential role in maintaining the stability of the bank line and protecting soil and water [17, 135-137]. Therefore, understanding the complexity and spatiotemporal dynamics of soil nutrient cycling processes in reservoir's buffer strips with different land-use types is an important task in assessing reservoir habitat quality and future ecological development [11, 120, 138, 139].

In this study, we found the soil TC and TN content in woodland buffer strips were significantly greater than in grassland and abandoned cropland, indicating that adopted agroforest ecosystems in the reservoir's buffer strips had better soil carbon sequestration potential, which was consistent with the literature of Cole et al., (2020) and Hazlett et al., (2005) [5, 14]. The woodland in this study adopted agroforest ecosystems, which have been widely proven to be suitable for development in buffer strips, maximized the resolution of cost-benefit conflict, and the synergistic effect of deep and submerged root systems as well as the efficient use of light pattern indicated that the adopted agroforest ecosystem in reservoir's buffer strips can significantly improve soil carbon stock chage [1, 2, 38]. It was noteworthy that the higher TC and TN in woodland buffer strips mainly came from the contribution at W2, which may be explained in two ways. First, due to the abundance of organic matter in the surface layer of woodland soil, the raised water level due to impoundment may have accelerated the release of nutrients from the surface litter. Second, the transfer of nutrients from high-elevation areas to watersheds with surface

runoff may have created a hotspot of carbon and nitrogen enrichment in the waterward location [79, 140], which indicates that adopted agroforest ecosystems in the reservoir's buffer strips need to be paid attention on the management of the nearwater area. In contrast, soil TP was significantly lower in woodland than the other two land-use types, which may be mainly due to the higher carbon and nitrogen content in woodland accelerating the activation of organic phosphorus and promoting the loss of dissolved phosphorus [79, 141, 142], which mean that adopted agroforest ecosystems has a lower capacity to trapped phosphorous, this reminds us that the use of pesticides in agroforest ecosystems buffer strips should be highly cautious, where organic farming may be better suited, this may also be one of the reasons why numerous studies have shown that woodland is less capable of trapping P than grassland [84, 143, 144]. In addition, the soil BD and SWC in woodland was significantly lower than the other two land-use types, indicating that the adopted agroforest ecosystems in the reservoir's buffer strips can effectively reduce soil water content and increase surface infiltration, which was consistent with the result of Anderson et al., (2009) [3].

Carbon and nitrogen pools are important factors affecting the soil nutrient cycle, and the mineralization of carbon and nitrogen can influence the amounts of ammonium and nitrates in soils [145, 146]. Studies have shown that woodland vegetation can provide more deep organic matter than grassland, can effectively promote denitrification, and can have a stronger ability to reduce nitrates [146, 147]. However, in this study, woodland soils were characterized by low NH<sub>4</sub>-N and high NO<sub>3</sub>-N, which may because water impoundment provides an anaerobic environment for abandoned cropland and grassland, promoting denitrification and accelerating nitrate depletion. The higher porosity of

woodland and better diffusion of oxygen in air-filled soli pores can inhibit denitrification and hasten the accumulation of nitrate, which means that adopted agroforest ecosystems in the reservoir's buffer strips can reduce Greenhouse gas emission [148, 149].

Studies have shown that increases in MBC and MBN are related to the availability of soil carbon (C) and nitrogen (N) sinks and that the decomposition rate of organic matter determines the change level of microbial C and N [150, 151]. In this study, the MBC, MBN, and DOC contents of woodland soil were significantly higher than those of abandoned cropland and grassland, which indicated that the agroforest ecosystems soil has well microbial activity. This may be attributed to two explanations. First, woodland has a more advanced root structure than abandoned cropland and grassland protect the soil from erosion, and is more conducive to microbial biomass accumulation [152]. Second, woodland soil had a better aeration environment that was conducive to the activity of aerobic microorganisms and thus enhanced the accumulation of microbial biomass [153]. The higher MBC, MBN, and DOC content in the woodland at W20 also seem to support the above speculation.

Ecological ratio constitutes an intrinsic link between biogeochemistry and the structure and processes of food webs, and is the result of the interaction of multiple chemicals in the ecosystem, reflecting the ecosystem's nutrient use strategy [56, 154-156]. Soil C/N, and MBC/MBN are usually considered to be relatively stable without large range fluctuations and are important indicators of ecosystem stability [157]. C/N is an indicator of N mineralization capacity and reflects the decomposition of soil organic matter. Low C/N (<25) indicates that the rate of organic matter decomposition is greater than the rate of accumulation [158]. In this study, soil C/N of the woodland buffer strips

was significantly greater than the other two land-use types, which indicated that the adopted agroforest ecosystems in the reservoir buffer strips had higher soil organic matter retention capacity. In addition, the C/N of all land-use types was lower than 12, which indicated that the soil nutrient status in the reservoir's buffer strips with three land-use types were in an environmentally constrained condition [121, 141]; MBC/MBN is usually considered as an effective indicator of nitrogen availability, and the lower the MBC/MBN, the higher the nitrogen availability [159, 160]. The strong correlation relationship between stoichiometric ratio and NO<sub>3</sub>-N in this study also provided direct evidence. Meanwhile, the MBC/MBN of the grassland was significantly lower than the other two land-use types, which indicated higher soil nitrogen availability in grassland.

# 4.3.2. Variation of soil physicochemical properties at the distance from the watercourse in reservoir's buffer strips with three land-use types

The effect of distance from the watercourse on the riparian ecosystem is mediated through the underground water level, the higher the underground water level, the more easily the vegetation and soil above are in contacte with water, which in turn affects the soil chemical processes and vegetation survival strategies [55, 120, 161]. The effects due to hydraulic gradient are more clearly defined at large scales and are still limited at fine-scale [4, 78, 120]. da Silva et al., (2021) proposed that the distance from the watercourse and the gradient of nutrient variation were the most important factors influencing the assemblage structure of pteridophytes in the tropical forests at fine-distance scale (< 50m) [161]. In the present study, we found that most of the nutrients did not seem to have a clear pattern of variation at the fine distance scale. While,

the results from our PCA analysis showed that the soil physicochemical properties at 0m were relatively similar for three land-use types, which may be due to the fact that the soil at this location was directly inundated with water and was thus more influenced by the nutrient status of the overlying water [101]. Without considering 0 m, we found that some soil nutrients showed a significant trend at the distance scale. For example, soil TC and TN of three land-use types gradually decreased with increasing distance for watercourse which was consistent with the previous literature [58, 120], although this difference was not significant in abandoned cropland and grassland. Meanwhile, the effects of distance from watercourse on MBC, MBN, and NH<sub>4</sub>-N all reached significant levels (Two-*ANOVA*). The results of the RDA analysis also indicated that distance from the watercourse was an important factor affecting the reservoir's buffer strips at fine distance scale. All of the above results indicated that the distance from the watercourse is still a non-negligible factor on soil physicochemical properties at fine distance scale (especially for sheltering measures such as buffer strips).

### 4.3.3. Relationship between soil physicochemical properties and vegetation biodiversity in reservoir's buffer strips with three land-use types

Soil properties are the external integrated manifestation of the interaction processes of various biotic and abiotic factors, and changes in soil texture, microbial activity, stoichiometric characteristics, and vegetation community structure can have profound effects on soil properties [120, 162, 163]. Hu et al., (2022) confirmed that the soil and microbial biomass stoichiometric is influenced by plant communities and soil properties in the mangrove forest. Therefore, clarifying the relationship between soil

physicochemical properties and vegetation with different land-use types and revealing the key factors affecting soil properties can provide a basis for the management and restoration of adopted agrofoest ecosystem in the reservoir's buffer strips [120, 164]. In the present study, the effect of land-use types on soil properties was achieved indirectly through the soil stoichiometric ratio, which suggested that the reservoir's buffer strips with different land-use types keep the relative constancy of soil physicochemical properties by maintaining stable stoichiometric characteristics. The effect of distance from the watercourse on soil properties was moderated by the vegetation biodiversity indices, and although the trend of the vegetation biodiversity indices was not significant at the distance scale, the strong correlation between the vegetation biodiversity indices and soil physical properties (SWC, BD) indicated that changes in vegetation biodiversity due to hydraulic gradient modulated the variation of soil properties in the reservoir's buffer strips. The results of the RDA analysis also showed that the Simpson index and distance from the watercourse were the two major factors influencing soil properties.

In addition, without considering the above-mentioned 0m, there seemed to be a different mechanism for the changing pattern of soil physicochemical properties in the reservoir's buffer strips with three land-use types, and the results of the PCA analysis showed that the woodland has a strong correlation with most of the soil chemical property parameters, microbial activity, and stoichiometric ratio characteristic, while the abandoned cropland and grassland had strong correlation with the soil physical properties and vegetation biodiversity indices, which may be due to the fact that the soil of woodland maintains relative well soil aeration, promoting soil microbial activity and accelerating nutrient turnover [169]. In contrast, abandoned cropland and grassland have

been subjected to the influence of water impoundment, their soil pores have been filled with water, their soil aggregates have become fragmented, and soil physical properties have become the dominant factor affecting soil nutrient status [155, 170].

Combining the above results, we can speculate that the existing buffer strips width of abandoned cropland and grassland do not seem to meet the needs of ecological restoration and should be appropriately widened. This was also in line with the proposal by Cole et al., (2020) that the width of riparian buffer strips should be set flexibly according to the land use [5], which also indicated that the adopted agroforest ecosystems can shorten the reservoir's buffer strips width setting while ensuring ecological benefits.

Simultaneously, combining the results of RDA and PLSM analysis indicated that MBN was a main factor affecting soil properties, and both land-use type and distance from the watercourse directly affect soil microbial activity. So microbial activity is an important indicator of soil physicochemical properties in the reservoir's buffer strips with different land-use types, which was consistent with the result of Ran et al., (2022) in the Three Gorges Reservoir riparian zone with different land-use [155].

#### 4.4. Conclusion

Soils are not only an important repository of ecosystem biodiversity but provide a wide range of other functions (e.g. carbon cycle, waste decomposition) and services (e.g. climate regulation, pathogen resistance), as well [171]. Understanding the complex dynamics of soil nutrient cycling processes, and thus characterizing the extent to which various potential soil indicators change in the absence of restoration, and the magnitude at which these changes occur, can help determine which indicators may be most useful

for effective monitoring of reservoir's buffer strips in the future [11].

In this study, we reiterated the critical role of distance from the watercourse on the heterogeneity of soil physicochemical properties in the buffer strips, both at large and fine distance scales. Adopted agroforest ecosystems have obvious advantages over other land-use types in maintaining soil microbial activity, and promoting soil carbon sequestration potential, reducing Greenhouse gas emissions. Therefore, adopted agroforest ecosystems in the reservoir's buffer strips have strong operability and showed obvious environmental performance advantages. Meanwhile, the width setting of the reservoir's buffer strips should be appropriately increased (>20m) in the case of restoration based on grassland and abandoned cropland. Soil microbial activity can be used as an important indicator for monitoring the soil nutrient status of different land-use types in the reservoir's buffer strips. However, these results were based on short-term preliminary investigations, and long-term monitoring of the coupling and decoupling mechanisms between soil physicochemical properties and vegetation needs to be carried out.

#### **CHAPTER 5**

### THE CHARACTERISTIC OF SOIL AGGREGATES STABILITY AND ASSOCIALTED NUTRIENT DISTRIBUTION FOR ADOPTED AGROFOREST ECOSYSTEMS IN RESERVOIR'S BUFFER STRIPS

#### 5.1. Statistical analysis

Mean weight diameter (MWD,  $\mu$ m) is used to evaluate soil aggregates stability, and we calculate MWD using the formula in Kemper and Rosenau (1986) [172]:

$$MWD = \sum_{i=1}^{3} X_i \times M_i$$

where *Xi* is the mean diameter of the *ith* particle ( $\mu$ m), *M<sub>i</sub>* is the mass proportion of ith size fractions in aggregates (%), and we calculate organic C pool (OCP, g/m2) in soil particle size fractions using the formula in Yang et al., (2007) [173]:

$$OCP = \sum_{i=1}^{3} (N_i \times OC_i) \times B_d \times H \times 10$$

where  $N_i$  is the mass proportion of the *i*th size fraction in whole soils (%),  $OC_i$  is the organic C content in the *i*th size fraction (g kg-1),  $B_d$  is the soil bulk density (g cm-3), and H is the soil thickness (cm).

All variables were tested for normality using the *Shapiro-Wilk* statistic, and those that did not conform to a normal distribution were transformed using the natural logarithm. We analyzed differences in soil particle size fractions, aggregates stability, particle-size associated carbon and nitrogen, and soil properties among different sampling sites using analysis of variance (*ANOVA*) with a least-significant test (*LSD*) post-hoc test (p < 0.05). Redundancy analysis (RDA) was conducted to explore the multivariate correlation between soil particle size fractions, MWD, and soil physicochemical properties using soil physicochemical properties as the environmental variables. RDA was also conducted to explore the multivariate correlation between soil particles, their associated nutrients, and soil physicochemical properties using soil physicochemical properties as the environmental variables. Stepwise multiple regression analysis was used to examine the linear relationship between soil particle size fractions, soil physicochemical properties, and particle-size associated nutrients; the partial regression sums of squares were used to determine the significance of each retention variable. Structural equation models (SEM) between land-use types, distance from the watercourse, sand particle-size, silt particle size, and clay particle size associated nutrients, soil microbial activity, and soil chemical properties were constructed using a partial least squares path model (PLS-PM) (soil physical properties were not included due to lack of BD at 0 m). All statistical analysis and plotting were executed in R (version 4.0.3).

#### 5.2. Results

The predominant soil particle size fractions in abandoned cropland and grassland is silt particle (42.08 to 75.43%), and the predominant soil particle size fraction in woodland is sand particle (29.37 to 80.26%), followed by silt particle (7.38 to 62.86%) (Table 5.1). We classify the soil texture of abandoned cropland and grassland as silt loam and woodland as sandy loam. Our MWD calculations for different land-use types showed that woodland > abandoned cropland > grassland, with woodland occupying a significantly larger area than the other two land-use types which indicated that adopted agroforest ecosystems in the reservoir's buffer strips have stronger aggregates stability, and our OCP calculations for different land-use types showed W20 > abandoned cropland > grassland > W2, and the OCP content of W20 was significantly greater than that at all other sites (Table 5.1).

Table 5.1

	Sand(53-2000µm)	Silt(2-53µm)	Clay(<2µm)	MWD(µm)	OCP(g/m <sup>2</sup> )
C0	21.39±9.60Babc	61.24±9.64Aab	17.36±6.84Ba	236.53±96.54bc	-
C2	13.70±3.79Babc	66.98±4.67Aa	19.31±8.23Ba	159.21±40.00c	3226.31±453.67b
C20	23.07±1.77Babc	58.65±2.67Aab	18.28±1.54Ba	253.01±17.55bc	2577.61±291.02bc
G0	9.32±2.10Bbc	68.46±4.28Aa	22.22±2.58Ba	114.58±20.48c	-
G2	7.57±1.28Cc	68.68±2.93Aa	23.75±2.89Ba	96.74±12.99c	2382.44±283.78bc
G20	7.70±2.79Cc	73.70±5.33Aa	18.60±7.99Ba	99.44±30.01c	2012.17±155.91bc
W0	55.00±12.03Aa	26.54±12.05Ac	18.46±1.46Ba	572.00±120.24a	-
W2	46.84±16.72Aabc	39.31±15.56Abc	13.86±5.90Ba	491.65±167.60ab	1685.19±149.89c
W20	51.49±7.49Aab	38.41±6.65Abc	10.10±0.97Ba	539.14±75.11a	4468.07±641.82a

Mean weight diameter (MWD), Organic C pool (OCP), and particle size fractions.

Different land-use types have different amounts of carbon, and carbon content varies depending on soil particle size. We found that the carbon content in grassland and abandoned cropland was the lowest in silt particles, which was significantly lower than that in clay particles at the C20, G0, G2, and G20 sites. We observed that the carbon content of each particle increased with the decrease in particle size in the woodland. In addition, we found that the carbon content in clay particles at W2 and W20 was significantly higher than that of sand, and the highest carbon content in sand particles was found at C2 (24.26 g/kg) and the lowest at W2 (6.40 g/kg), which mean that adopted agroforest ecosystems in reservoir's buffer strips can prevent carbon loss and have high carbon stock change. We saw the highest carbon content in silt and clay particles at W20

(23.56 g/kg and 28.62 g/kg, respectively), which was significantly higher than the other sites (Figure 5.1).

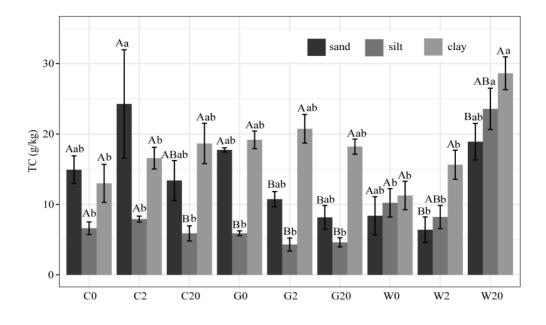


Figure 5.1. Characteristics of soil particle-size associated carbon distribution.

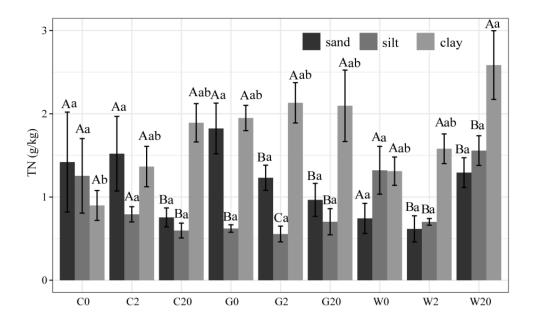


Figure 5.2 Characteristics of soil particle-size associated nitrogen distribution.

The distribution characteristics of soil particle-size-associated nitrogen on different land-use types and sites show a similar trend to soil particle-size-associated carbon (Figure 5.2). Several studies have shown that carbon and nitrogen in soil are highly coupled [118, 174, 175], therefore, this study focuses on the distribution of soil particle-size associated carbon.

Using RDA analysis, we explored the relationship between soil particle size fractions, MWD, and soil physicochemical properties of different land-use types and showed that the interpretation of soil physicochemical properties, in terms of soil particle size fractions and MWD, was mainly concentrated on the first axis of RDA, reaching 34.36%. We found that pH, STC, STN, DOC, MBC, and MBN were positively correlated with sand particles and MWD; soil physicochemical properties indicators, sand particles, and MWD were mainly concentrated in the area where the woodland samples were distributed and that silt particle and clay particle were mainly concentrated in the areas where the abandoned cropland samples were distributed (Figure 5.3).

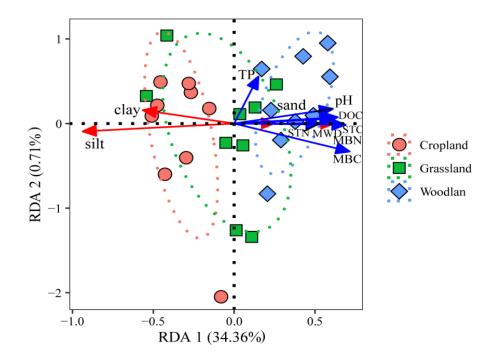


Figure 5.3 Redundancy analysis (RDA) for the soil particle size fractions and MWD

with soil physicochemical properties as explaining variables.

Similarly, we also explored the relationship between soil particle-size associated carbon, nitrogen distribution, and soil physicochemical properties in different land-use types using RDA. We found that the degree of explanation is 19.1% on the first axis and 17.48% on the second axis of RDA. pH, STC, STN, DOC, and MBC were positively correlated with TC2 (silt particle-size-associated carbon) and TN2 (silt particle-size associated nitrogen) and were mainly concentrated in the areas where the woodland samples were distributed; TC3 (clay particle-size associated carbon) and TN3 (clay particle-size associated nitrogen) were concentrated in the areas where the abandoned cropland samples were distributed, and TC1 (sand particle-size associated carbon) and TN1 (sand particle-size associated nitrogen) were concentrated in the areas where the grassland samples were distributed (Figure 5.4).

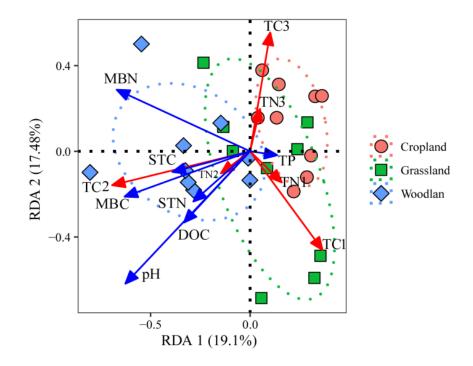


Figure 5.4 RDA for the soil particle-size associated carbon and nitrogen with soil

physicochemical properties as explaining variables.

We found that MWD and particle size fractions were closely related to the corresponding soil physicochemical properties and particle-size-associated nutrients (Table 5.2). Specifically, DOC positively correlated with MWD, while MBC negatively correlated with MWD. In addition, MBC positively correlated with silt particles but negatively correlated with sand particles. In the multiple regression equation of sand particle, silt particle, and MWD, TC2 was the main variable that influenced the equation (Table 5.2).

Table 5.2

Aggregate properties	Linear regression model with partial correlation coefficient			
Sand	21.120-15.751TN1(21.15)+5.102TC2(49.27)-3.384TC3(16.04)+21.925T N3(7.76)+44.428STP(5.98)+1.135DOC(1.87)-0.178MBC(10.90)	0.8618		
Silt	37.597+13.974TN1(10.84)-4.359TC2(25.26)+0.903TC3(2.87)-25.209ST P(1.91)+0.169MBC(6.51)	0.7044		
Clay	45.08-9.665TN2(10.79)+0.873TC3(2.78)-13.028TN3(5.83)-0.482STC(4. 68)-15.571STP(1.86)	0.4556		
MWD	230.063-157.679TN1(21.25)+51.162TC2(49.70)-34.269TC3(16.49)+222. 819TN3(8.04)+447.551STP(6.09)+11.472DOC(1.91)-1.776MBC(10.90)	0.8632		

Multiple linear regression analysis of particle size fractions, MWD, particle-size

A more in-depth analysis using PLS-PM and showing the direct and indirect effect of land-use types and distance from the watercourse on sand, silt, and clay particle-size associated nutrients, microbial activity, and soil chemical properties is needed (Figure 5.5). Distance from the watercourse on sand particle-size-associated nutrients had the highest path coefficients (0.78, p < 0.0001). The effect of silt particle-size associated nutrient and clay particle-size-associated nutrients on microbial activity had relatively

associated nutrients, and soil physicochemical properties.

high path coefficients (-0.59, p = 0.019; 0.69, p = 0.036). The goodness of fit (GOF) was 0.49, which indicated that the model had relatively good predictive power.

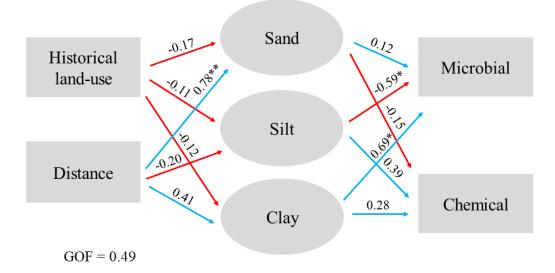


Figure 5.5 Causal inference diagram based on PLS-PM..

#### **5.3.** Discussion

The establishment of the reservoir's buffer strips is a management measure that has been widely promoted worldwide as an effective way to reduce pollution and improve water quality [21]. In this study, different land-use types in the reservoir's buffer strips experienced complex dynamics in soil aggregate stability, particle-size associated nutrient distribution characteristics, and geochemical processes after water impoundment, showing distinct response mechanisms at different distances from the watercourse.

# 5.3.1. Soil aggregates stability characteristics of different land-use types in reservoir's buffer strips

In this study, we classified the soil texture of the woodland as sandy loam and the

abandoned cropland and grassland as silt loam. We found that the MWD of woodland was significantly greater than that of the other two land-use types, indicating that adopted agroforest ecosystems in the reservoir's buffer strips had greater soil aggregate stability and were better able to resist the effects of hydraulic erosion [176, 177].

Many studies have reported the response mechanisms of soil particle size fractions and soil aggregate stability in the reservoir riparian zone during seasonal dynamics and long-term alternating dry-wet cycles [92, 178, 179]. Therefore, the process of soil aggregate stability in response to inundation is still controversial. Sarah & Rodeh, (2004) suggested that soil would gradually transition toward a typical nature "steady state" during wetting, and soil aggregates stability would increase [180]. The more common view is that after repeated dry-wet cycles, the shrinkage and expansion of the soil due to hydrological stress leads to a stabilization of the particle size distribution and a decrease in aggregate stability [99]. E.g. Nsabimana et al., (2020) studied the impacts of water level fluctuations on soil aggregates stability in the Three Gorges Reservoir and found that MWD decreased gradually along the elevation [140]. The reasons for these two differences may be closely related to the study region and soil-submerged conditions. In this study, both grassland and woodland showed a tendency for MWD at 0 m to be greater than at 2 m and 20 m. The soil at 0 m had been submerged since water impoundment and was rarely exposed, which suggests that short-term wetting facilitates soil aggregates stability, and this assumption is consistent with the findings of Denef et al., (2001), Totsche et al., (2018), and others [86,95]. While, the MWD at 0m of abandoned cropland is greater than 2m and less than 20m, which may be related to less aboveground vegetation coverage in this area [181, 182].

MWD at 2 m is lower than at 0 m and 20 m for the different land-use types, suggesting that water impoundment reduces soil aggregate stability in the waterward location of the reservoir's buffer strips [140,183]. This finding may be mainly due to the fact that 2 m is closer to the watercourses where the soil is frequently in alternating dry-wet cycles caused by groundwater table elevation and surface runoff, which makes the soil at this location more susceptible to disintegration and thus reduces soil aggregates stability [91, 162].

In summary, the reservoir's buffer strips with different land-use types showed significant differences in soil aggregate stability at fine distance scales after reservoir impoundment because of the dry-wet cycles and water submerge conditions caused by different distances from the watercourse.

# 5.3.2. Particle-size associated nutrient distribution of different land-use types in reservoir's buffer strips

Soil organic matter (SOM) is an essential cementing substance in soil aggregates and determines the formation of water-stable aggregates, and the size and mass percentage of soil particles significantly affect the size and quantity of soil aggregates and their binding to organic matter [165, 184]. Soil organic carbon obtained through soil particle size grouping is an effective entry point to understand further the relationship between the decomposition transformation and stability of the SOM and the location and state in which it exists [185, 186].

In this study, without considering 0m site, we observed that the carbon content in sand particles was significantly lower than that in silt particles of both grassland and woodland, nevertheless, the woodland did not show such tendency, and silt particle size associated with carbon was higher than in sand particle size, which indicated that the agroforest ecosystems can prevent carbon loss under water submerge condition. In addition, soil particle-size associated carbon distribution at 0m may influenced by organic matter carried by overlying water which is consistent with the results of Chapter 4 [101]. Although the carbon content in sand particles at W0 and W2 of the abandoned cropland was greater than that of the clay particle, the difference was not statistically significant probably because the carbon content of the soil macro-aggregate increased during previous cultivation [13, 187]. In general, in the restored soil types, the organic carbon and nitrogen that can be bound by the soil macro-aggregate was greater than that of the micro-aggregate [93, 188]. We speculated that the loss of carbon in the sand particle of different land-use types in the reservoir's buffer strips occurred to different degrees after water impoundment possibly because the sand particle was the main source of the soil's active carbon pool and, therefore, more vulnerable to the risk of loss by external erosion such as water inundation [147].

The response of soil particle-size-associated carbon to external erosion, such as inundation, is accompanied by fragmentation and regeneration of different particle size fractions, and the direction of evolution depends on which process is dominant [86, 189]. Soil organic matter adsorption on clay particles is an important determinant of soil organic matter stability, and fine silt-clay particles ( $< 20\mu m$ ) have the maximum capacity to bind soil organic carbon in different soil types. Therefore, one of the main factors in the physical conservation of soil organic matter is its ability to bind with fine silt-clay fractions [190, 191]. Among them, the silt particles are the intermediaries of soil organic

carbon cycle, and when disturbed by external forces, the microorganisms attached to the silt particle can use the organic carbon isolated by the silt particle to accelerate the nutrient turnover between different particle sizes [7, 95, 192]. In this study, the silt particle-size associated carbon content was generally low except in W20, while the silt particle content in woodland was the lowest, which indicated that soil silt particles in agroforest ecosystems had a high carbon binding capacity. In addition, the soil silt particle-size associated carbon content of woodland showed greater than sand particle-size associated carbon content and abandoned cropland and grassland showed the opposite trend. This is possibly because the woodland had a more abundant root system, which enhanced soil aeration, and there could be two different response mechanisms at different distances from the watercourse in the woodland because of the effect of the water table. In the waterward region, the soil condition was affected by water dam-triggered flood intensity, nutrient turnover between soil particles was accelerated, and the silt particle-size associated carbon content was reduced. While in the landward region, which was less stressed by water inundation, clay particle was still able to provide good physical protection for the soil organic matter. In contrast, both abandoned cropland and grassland were affected by water storage to varying degrees, and nutrient turnover between different particle sizes was accelerated. Extensive studies have shown that the width of the reservoir's buffer strips should vary according to different land-use types [73, 193]. Therefore, we speculated that the 20 m we set for abandoned cropland and grassland may not completely cover the dam-triggered flooding impact area, and the width setting for the abandoned cropland and grassland buffer strips should be expanded for practical restoration, this result is consistent with Chapter 4.

In this study, we also observed that soil total carbon (STC) was significantly higher in W2 than in W20 probably because, after reservoir impoundment, ample organic matter in the woodland migrated from upper to lower areas with surface runoff, and the fluctuation in water level also brought a large amount of organic matter to the bank, creating a hot spot for carbon accumulation in the waterward region [140, 194]. However, the particle-sizes associated carbon content was significantly higher in W20 than in W2, and there was apparently a large margin in between, which was not significantly observed in other historical land-use types. A possible explanation is that after reservoir impoundment, the release of organic matter exists in the form of inorganic carbon, and the carbonate forms precipitate with some metal ions in the water, which are stored on the surface of the sediment [195]. This situation also explains the low OCP we calculated in W2, which suggested that the use of particle sizes grading to estimate carbon stock change in the reservoir buffer strips may result in an underestimation of agroforest ecosystems' carbon sequestration.

Taken together, the reservoir's buffer strips of different land-use types all underwent corresponding changes in the particle-size associated carbon distribution characteristics after reservoir impoundment mainly due to the turnover properties of different soil particles combined with organic carbon.

# 5.3.3. Relationship between soil particle fractions and chemical properties in historical land-use types in reservoir's buffer strips

In the process of reservoir buffer strip evolution and restoration, soil texture is affected by internal factors of the soil parent material and external factors such as soil chemical composition, climate, water, and vegetation [196, 197]. Dam-triggered flooding intensity also determines the trend of soil texture and nutrients [174, 198]. The dynamics of nutrients (e.g., nitrogen, phosphorus, and potassium) can indirectly affect the stability of the soil aggregates and the associated carbon distribution patterns [199].

Reviewing the relationship between soil aggregate stability and SOM, Six and Paustian (2014) stated that, for a long time, SOM physical fractionation emphasized the influence of soil substrate on SOM pool size and dynamics, but did not have a good correlation with soil chemical properties. In this study, we found that sand particles, silt particles, and MWD were well correlated with soil physicochemical properties and particle sizes associated with carbon content ( $R^2=0.86$ ,  $R^2=0.74$ ,  $R^2=0.86$ ), and TC2 was the most significant determinant. The results of RDA and PLS-PM analyses showed that the distribution of silt particle-size-associated carbon was closely related to soil microbial activity, which suggested that soil microbial activity was an important factor influencing the distribution of soil particle-size-associated carbon content in the reservoir's buffer strips. It is well known that soil microorganisms play an essential role in the formation, stabilization, and destruction of soil aggregates [200]. Different microbial communities prefer to colonize different soil particle size fractions and are adapted to the physical and chemical properties of the soil [193, 201]. This difference in preference may be due to the transition from aerobic to anaerobic soil microbial metabolism, which evolved to adapt in specific environments for different land-use types in the reservoir's buffer strips after reservoir impoundment where microbial activity becomes a major limiting factor for soil ecology function [202]. Combined with the important role of nutrient turnover in silt fraction, we can assert that reservoir impoundment accelerates the turnover of silt

particle-size associated nutrients in soils of different land-use types in the reservoir's buffer strips and this process may be mediated mainly by microbial activity.

It is worth noting that the sampling time in this study was carried out two months after the reservoir reached its baseline (normal level), and only one sampling was carried out. The limitation of this study is that it can only represent the soil characteristics after short-term inundation. The long-term evolution mechanism of soil particle-size associated carbon cannot be deeply explored. Secondly, due to the patchy distribution characteristics of historical land-use types around the reservoir and the time limit for experimenting, it is difficult to obtain soil background value before water impoundment, and there is a lack of effective comparison.

#### **5.4.** Conclusions

The stability of soil aggregates and organic matter distribution characteristics are key mechanisms in understanding the evolution of the environment in and around reservoirs; a better ecological understanding can aid in the management of reservoir buffer strips in general [21]. In this study, we found that soil aggregate stability and particle-size associated carbon distribution characteristics of adopted agroforest ecosystems in the reservoir's buffer strips showed different response mechanisms after reservoir impoundment, which were mainly caused by the dry-wet cycle and water submerge conditions at different distances from the watercourese and soil associated carbon cycle mechanisms of different particle size fractions.

The impact of dam construction on reservoir environments is long-lasting and far-reaching [203]; different land-use types around a reservoir gradually transition to an

aquatic ecosystem, so this study is an important guide to designing and restoring the reservoir's buffer strips. In future reservoir buffer strip management, different protection measures and width settings should be adopted for different land-use types, and high priority should be given to the protection of waterward regions.

As for adopted agroforest ecosystems in the reservoir's buffer strips, we can see that it can improve the soil aggreages stability, and carbon stock change, prevent carbon loss, and maintain the stability of the shoreline. Especially the TC2 (silt-associated carbon content) was the importance for the carbon cycle for adopted agroforest ecosystems in the reservoir's buffer strips. Also, as the influence of water level fluctuation in nearwater area, we need to pay attention to this region.

We focused on soil aggregate stability and associated organic matter distribution characteristics of different land-use types in the reservoir's buffer strips after reservoir impoundment, but the long-term mechanism of soil ecology with seasonal drainage and storage warrants further study.

#### **CHAPTER 6**

### THE CHARACTERISTIC OF MICROBIAL COMMUNITY COMPOSITION AND ASSEMBLY MACHANISM FOR ADOPTED AGROFOREST ECOSYSTEMS IN RESERVOIR'S BUFFER STRIPS

#### 6.1. Statistical analysis

Before analysis, ASVs of all sequenced samples were rarefied to 13000 (minimum sequence depth) to ensure consistency. All data were tested by the Shapiro-Wilk test to check data normality. Microbial community diversity indices were calculated (Simpson index, Shannon index, Richness index, Chao1, PD whole tree index) using the Vegan package in R. Using PcoA to show the  $\beta$  diversity of microbial community for different land-use types based on the bray-Curtis distance matrix, Weighted UniFrac and Unweighted UniFrac distance matrices separately, significance between the subgroups was calculated using PERMANOVA after 999 permutation test. The relationship between soil physicochemical properties, vegetation diversity, and microbial communities was exhibited for different land-use types using CCA, and the main factor retained was screened using *ordiR2step* in the RDA package and combined with prior experience, the rdacca.hp [204] package was used to decompose each factor's explanatory degree. The NTI values were calculated the standardized effect size measure of mean nearest taxon distance (SES.MNTD) with the ses.mntd function in the *Picante* package of R to evaluate the phylogenetic status of different land-use types and sites. The soil physicochemical properties and vegetation diversity index were decomposed into soil physical properties (soil water content, bulk density), fast turnover chemical properties (NH<sub>4</sub>-N, NO<sub>3</sub>-N,

DOC, MBC, MBN), slow turnover chemical properties (TC, TN, TP), Soil texture (sand content, silt content, clay content), Soil particle size associated nutrient distribution (TC1, TN1, TC2, TN2, TC3, TN3), stoichiometric ratio properties (MBC/MBN, C/N) and plant diversity (Shannon diversity index, Richness diversity index). The mantel.test was used to calculate the relationship and significance between each decomposition attribute and the top 10 abundance microbial taxa at the phylum level with the *LinkET* package. The ggClusterNet package was used to display the co-occurrence network and calculate the topological parameters, retaining species that occurred simultaneously in at least three samples with thresholds set as  $r \ge 0.8$  and  $p \le 0.01$ . The effect of land-use type and distance from the watercourse on microbial diversity indices were calculated using two-way-*ANOVA*. All analyses and visualization of graphs were performed using R studio 4.1.3 version.

#### 6.2. Results

The microbial community diversity indices between different land-use types in the reservoir's buffer strips were not observed as significant, while the woodland showed higher than the other two land-use types, which indicated that adopted agroforest ecosystems can maintain high microbial biodiversity. However, from the point of distance scale, the microbial community diversity indices of abandoned cropland and woodland showed significantly greater at 2m than 20m (Figure 6.1.), indicating that the microbial community structure and phylogeny of these two land-use types differed significantly between water-ward and land-ward areas. The results of two-way-*ANOVA* also indicated that distance from the watercourse and interaction effects were the main

factors affecting the diversity index of microbial communities in the reservoir's buffer strips (Table 6.1).

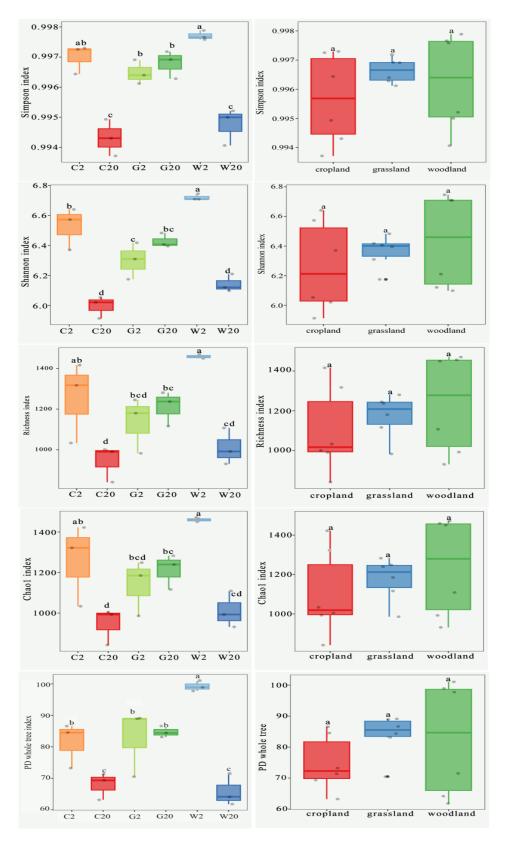


Figure 6.1 Varibation of microbial community diversity indices.

	Land-use				distance			Land-use×distance	
	df	F	Р	df	F	Р	df	F	Р
Simpson	2	6.331	0.013*	1	61.518	0.001**	2	21.501	0.001**
Shannon	2	5.725	0.018*	1	62.795	0.001**	2	30.383	0.001**
Richness	2	1.999	0.178	1	17.238	0.001**	2	8.130	0.006**
Chao1	2	1.908	0.191	1	17.042	0.001**	2	7.909	0.006**
PD_whole_tree	2	4.044	0.045*	1	28.153	0.001**	2	13.042	0.001**

The correlation and interaction of microbial community indices between land-use

types and distance from watercourse (k=6, n=3)

The results of PcoA analysis showed that the  $\beta$  diversity of microbial communities of different land-use types in the reservoir's buffer strips showed significant differences in terms of sample similarity distance and phylogeny (p = 0.001). Based on Bray-curtis similarity, Weighted UniFrac and Unweighted UniFrac distance matrices explained 57.25%, 70.21%, and 49.95% of the microbial community for different land-use types, respectively. The PcoA results based on different distance matrices all showed that the distribution of microbial communities in the grassland of the different sites were clustered together. In contrast, the microbial communities of abandoned cropland and woodland at 2m and 20m were distributed on both sides of different axes, this indicated that adopted agroforest ecosystems in the reservoir's buffer strips have distinct micrbial community distribuion pattern at distance scale (Figure 6.2, Figure 6.3).

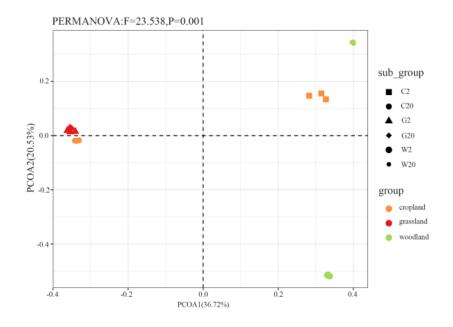


Figure 6.2 Principal coordinates analysis (PCoA) base on Bray-curtis dissimilarities

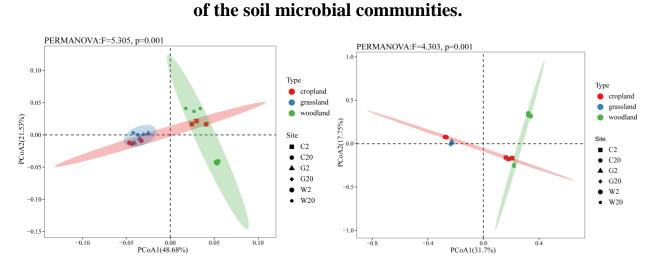


Figure 6.3 Principal coordinates analysis (PCoA) base on Weighted UniFract (Left) and Unweight UniFract (Right) dissimilarities of the soil microbial communities.

The species composition of microbial communities in different land-use types of reservoir buffer strips was mainly dominated by the phylum with top 10 abundance, accounting for 94.3% on average. Grassland showed a predominance of *Proteobacteria* (27.1%, 25.1%) in both G2 and G20, followed by *Acidobacteriota* (20.6%, 22.1%), *Chloroflexi* (18.8%, 19.4%), *Actinobacteriota* (13.7%, 14.4%). C2 showed a

predominance of *Proteobacteria* (26.6%), followed by *Actinobacteriota* (24.5%), *Acidobacteriota* (18.3%), and *Chloroflexi* (7.25%). C20 showed a predominance of *Acidobacteriota* (24.1%), followed by *Proteobacteria* (22.7%), *Chloroflexi* (17.0%), and *Actinobacteriota* (14.6%). W2 showed a predominance of *Actinobacteriota* (25.3%), followed by *Acidobacteriota* (17.6%), *Chloroflexi* (17.4%), *Proteobacteria* (14%). W20 showed a predominance of *Actinobacteriota* (25.3%), followed by *Proteobacteria* (24.7%), *Acidobacteriota* (18.9%), *Chloroflexi* (12.2%) (Figure 6.4).

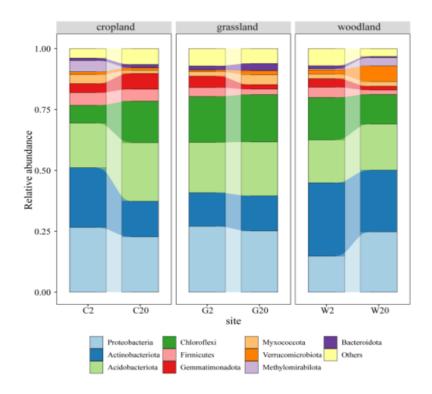


Figure 6.4 Column accumulation diagram show the composition pattern of bacterial community in the relative abundance of predominant taxa (phylum level).

The abundance of *Chloroflexi* was significantly greater in grassland than in abandoned cropland. The abundance of *Bacteroidota* was significantly greater in grassland than in abandoned cropland and woodland. The abundance of *Actinobacteriota* and *Methylomirabilota* were significantly greater in woodland than in grassland (Figure

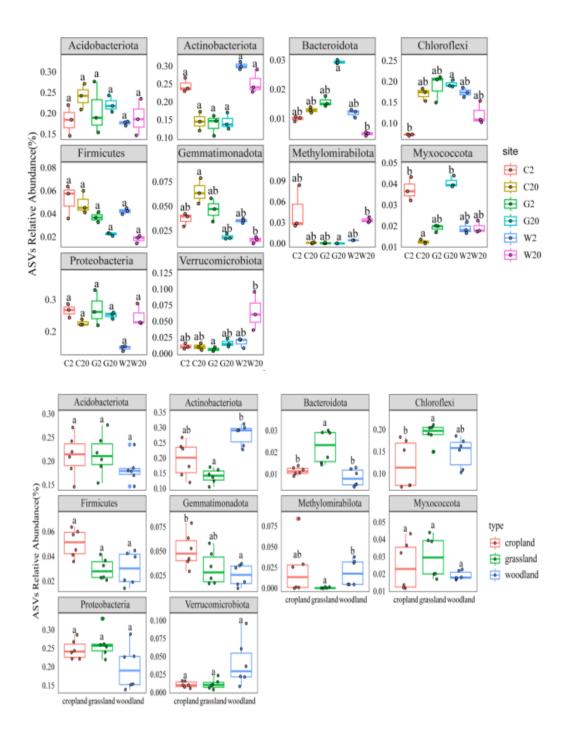


Figure 6.5 Differences of the top 10 abundance microbial community taxa (family level).

The results of CCA analysis showed that the screened soil physicochemical parameters and vegetation diversity index explained 7.2% of microbial community

distribution in the first axis and 6.7% in the second axis (Figure 6.6., Left). Among them, BD, Soil moisture, and NH<sub>4</sub>-N were positively correlated with grassland and C20, and negatively correlated with TC, Simpson, pH, and DOC. TC was positively correlated with W2. TC had the greatest explanatory degree for the distribution of soil microbial communities in the reservoir's buffer strips (Figure 6.6, Right).

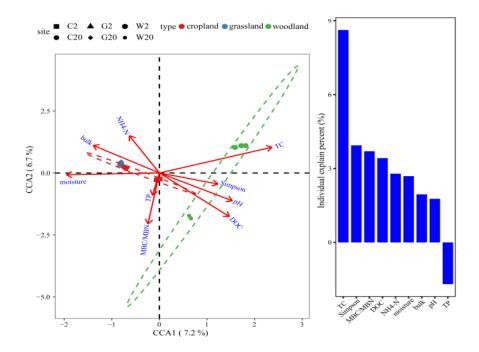


Figure 6.6 Canonical Correspondence Analysis (CCA) to display the relationship between soil properties, plant diversity and microbial communities (Left). The explanation percents of the screened individual factors to microbial community

#### (Right).

Soil physical properties were strongly correlated with *Actinobacteriota* abundance and weakly correlated with *Verrucomicrobiata* and *Methylomirabilota* abundance. Soil show turnover chemical properties were strongly correlated with *Actinobacteriota* abundance. Soil fast turnover chemical properties were strongly correlated with *Firmicutes* and *Verrucomicrobiota* abundance. Soil texture was strongly correlated with *Actinobacteriota* abundance. Soil particle sizes associated nutrients distribution were strongly correlated with *Firmicutes* abundance and weakly correlated with *Chloroflexi* and *Verrucomicrobiata*. Soil stoichiometric ratio was strongly correlated with *Proteobacteria* and *Actinobacteriota* (Figure 6.7).

The abundance of *Proteobacteria* was positively correlated with *Acidobacteriota*, and negatively correlated with *Actinobacteriota* and *Methylomirabilota*. The abundance of *Actinobacteriota* was positively correlated with *Methylomirabilota*, and negatively correlated with *Chloroflexi* and *Bacteroidota*. The abundance of *Chloroflexi* was negatively correlated with *Methylomirabilota*, and positively correlated with *Bacteroidota* (Figure 6.7).

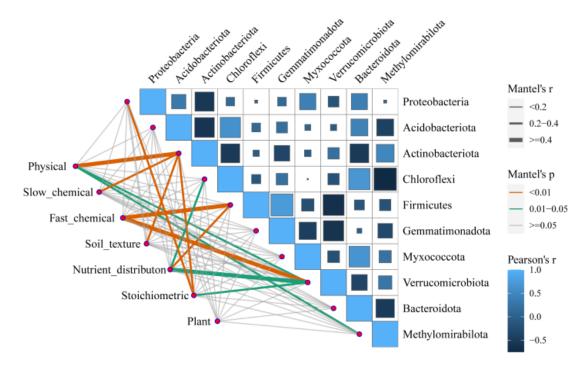


Figure 6.7 Correlation between the top 10 abundance microbial community taxa (phylum level) and soil properties, plant diversity indices.

In contrast to grassland, more ASVs were depleted in abandoned cropland, the top5 abundance classified at Phylum level were *FCPU426*, *Preteobacteria*, and

*Acidobacteriota*. In contrast to woodland, more ASVs were enriched in grassland, and the top 5 abundance classified at the Phylum level were *Acidobacteriota*, *Actinobacteriota* and *Proteobacteria*. In contrast to woodland, more ASVs also were enriched in abandoned cropland, and the top 5 abundance classified at Phylum level were *Acidobacteriota*, *Actinobacteriota*, *Proteobacteria*, *Chloroflexi*, which indicated that adopted agroforest ecosystems in reservoir's buffer strips have relative stability microbial community composition than the other two land-use types (Figure 6.8).

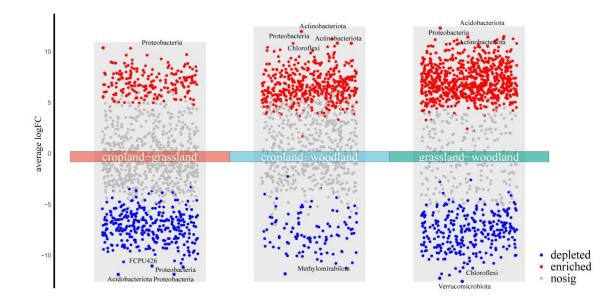


Figure 6.8 The volcano diagram of significantly difference microbial community taxa.

The co-occurrence network of different land-use types in the reservoir's buffer strips had obvious differences (Figure 6.9). The number of network nodes, positive links, connectance, and average degree are all shown as abandoned cropland > woodland > grassland. The average path length of grassland is significantly larger than the other two land-use types, which indicates that the network structure of abandoned cropland is more complex, and that of grassland is relatively simple. It is noteworthy that the number of negative links in woodland is larger than the other two land-use types, and the diameter and cluster coefficient are smaller than the other two land-use types, which indicates that adopted agroforest ecosystems in the reservoir's buffer strips have relatively stable network structure (Table 6.2). The centralization betweenness and centralization closeness of grassland were both significantly greater than the other two land-use types.

Table 6.2

	cropland	grassland	woodland
edges	4054	1206	2986
positive	3357	670	2114
negative	697	536	872
vertices	194	188	179
connectance	0.2165	0.0686	0.1874
Average degree	41.7938	12.8297	33.3631
Average path length	2.0476	4.3449	2.0994
diameter	6.5337	14.99437	5.5685
Clustering coefficient	0.6726	0.6427	0.5889
Centralization degree	0.1151	0.1025	0.1103
Centralization betweenness	0.0308	0.2197	0.0247
Centralization closeness	1.0668	1.5206	1.0823

**Co-occurrence network topology parameters** 

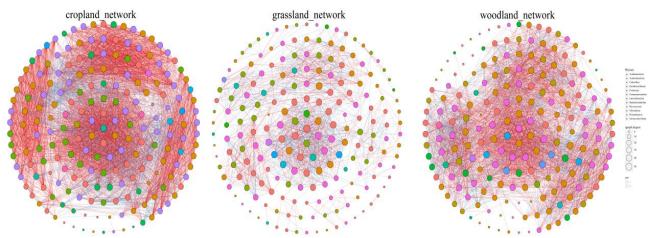


Figure 6.9 Co-occurrence network of bacterial community taxa.

The neutral community model was applied to explore the community assemblies of different land-use types in the reservoir's buffer strips. The community  $R^2$  explained by the bacterial taxa of grassland was 0.69, while only -0.138 and 0.03 for abandoned cropland and grassland, indicating that the neutral community model was not suitable for abandoned cropland and woodland (Figure 6.10). The metacommunity size multiplied by immigration (Nm) was significantly higher for the grassland than the other two land-use types, which indicated the greater contribution of stochastic processes in the grassland.

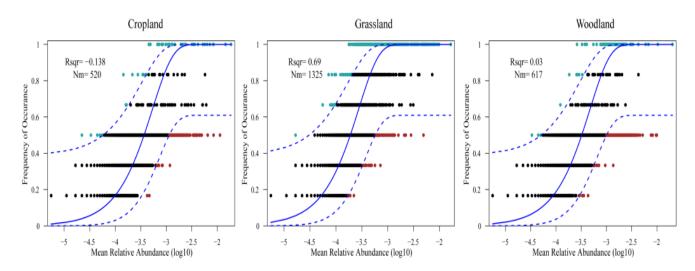


Figure 6.10 The relationship between occurrence frequencies and the relative

abundance of ASVs.

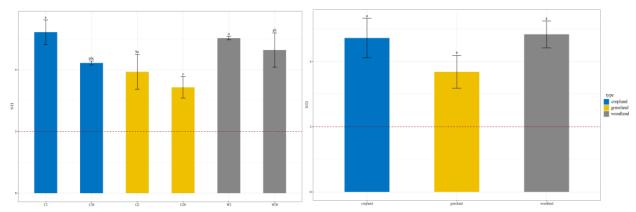


Figure 6.11 Histogram of NTI indicex for different land-use types (Right) and site (Left).

The NTI index of different land-use types in the reservoir's buffer strips showed that grassland was significantly smaller than abandoned cropland and woodland (Figure 6.11). The NTI index at G20 was the lowest and significantly smaller than W2, W20, and C2. The NTI index of different sites significantly deviated from zero, which indicated that the microbial community structure in the reservoir's buffer strips was dominant by the phylogenetic cluster, and was structured by environmental filtering.

Further analysis of microbial community assembly processes in different land-use types showed that the main process that dominated microbial community structure in abandoned cropland was dispersal limitation, followed by heterogeneous and homogeneous selection; the main process that dominated microbial community structure in grassland was homogeneous selection; the main processes dominated microbial community structure in woodland was homogeneous selection, followed by dispersal limitation (Figure 6.12).

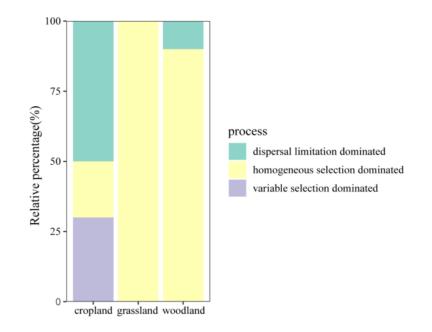


Figure 6.12 Assembly pattern of bacterial community.

Function predictions allowed for inferring differences in the functional characteristics of microbial communities in different habitats. In this study, the metabolism-related microbial community functions such as Metabolism of terpenoids and polyketides, Carbohydrate metabolism, Amino acid metabolism, and Energy metabolism were all in high abundance, which indicated that the soil microbial activities were intensive for different land-use types in the reservoir's buffer strips (Figure 6.13). Among them, the microbial community functional abundance of Cell motility and Signal transduction in woodland were significantly lower than the other land-use types; while Metabolism of terpenoids and polyketides, Carbohydrate metabolism, Amino acid terpenoids and polyketides were significantly higher than the other land-use types. The microbial communities functional abundance of Energy metabolism was significantly lower than the other land-use types. All of this indicated that adopted agroforest ecosystems in the reservoir's buffer strips have stronger metabolic activity to maintain the functional stability of the microbial community.

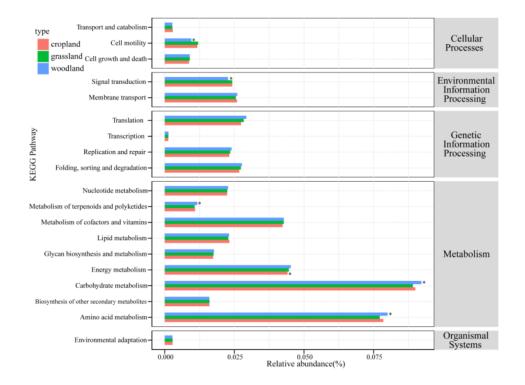


Figure 6.13 Histogram of predicted soil microbial community function.

#### 6.3. Discussion

Microbial communities are very sensitive to environmental changes and are shaped by a combination of biotic (vegetation, animals, etc.) and abiotic factors (pH, soil moisture, etc.). Soil microorganisms in the reservoir's buffer strips are not only threatened by the external hydrological stress but also by the internal microenvironment of the habitat [112, 126]. Differences in the vegetation type, nutrient uptake by root systems, and the quality of litter input due to land use, which in turn alter soil formation processes and nutrient cycling, indirectly affecting the composition and function of microbial taxa and community assembly mechanisms [114, 205]. Therefore, exploring the function and characteristics of soil microbial communities for different land-use types in the reservoir's buffer strips can help to comprehensively and deeply understand the response processes and mechanism of soil microorganisms in a highly complicated and variable reservoir shoreline environment.

## 6.3.1. Composition and diversity of microbial communities for different land-use types in reservoir's buffer strips

Microorganisms play essential roles in riparian ecosystems by maintaining environmental and ecological processes and nutrient transformation and cycling [206]. The most abundant phylum in our study included Proteobacteria, Acidobacteriota, Actinobacteria, and Chloroflexi which are also typical phyla in water-land ecotone ecosystems [105,207], although the relative abundances were different between land-use types and sites. We also found that taxa such as Actinobacteria, Chloroflexi in high abundance and Methylomirabilota, Gemmatimonadot in low abundance differed significantly between land-use types and sites, suggesting that different habitats in reservoir's buffer strips contain specific microbial taxa and that these taxa simultaneous indirectly reflect the functional distinctions among land-use types [152]. For example, previous studies have demonstrated that Actinobacteria and Chloroflexi can possess a competitive advantage relative to other phylum in soils with low nutrient availability [209]. Acidobacteria and Chloroflexi are the dominant decomposers of OM [210]. Further, we found that the functional activity of the microbial community related to material metabolism was higher in different land-use types in the reservoir's buffer strips, with woodland performing significantly better than the other land-use types, indicating that the alternating dry-wet condition in the reservoir's buffer strips significantly improved the metabolism function of the microbial community in response to the stress from the environment, while adopted agroforest ecosystems in reservoir's buffer strips showed

more activity.

We found no significant differences in microbial community diversity between land-use types, which is consistent with the results obtained from Three Gorge Reservoir with different habitats [207], suggesting that soil microorganisms of different land-use types in the reservoir's buffer strips have a certain adaptive capacity to the dramatically fluctuating hydrological environment. Similar results were observed by [208] in their study of the effect of short-term dry-rewet cycling on soil microbial community composition conducted in the laboratory. However, when the distance from the watercourse was taken into account, both woodland and abandoned cropland soil microbial community diversity exhibited greater in water-ward than land-ward, which is in line with the results observed by Ding et al., (2022) in wetland habitats of the Taihu Lake [152], and in contrast with the results for woodland and grassland, which may be due to the scale of sampling distances than ours'. Previous studies have demonstrated that moderate short-term disturbances increase the diversity of microbial communities and promote community realignment [211]. Yang et al., (2019) observed higher microbial community diversity in the Three Gorge reservoir riparian zone with medium flooding, suggesting that the marginal effects of the land-water interface would promote microbial community function more activity [106]. From the results of the Two-Way ANOVA, we also found that distance from the watercourse was the main factor influencing microbial community diversity, suggesting a noticeable scale effect on the distribution of microbial community diversity at fine distance scales in the reservoir's buffer strips. da Silva et al., (2021) observed similar assembly patterns of pteridophytes at the fine-distance scale in the riparian zone [161].

# 6.3.2. Major factors affecting the distribution of microbial community for different land-use types in reservoir's buffer strips

Numerous studies have shown that land use is an important factor driving soil microbial community structure and function, indirectly affecting it by modifying soil physicochemical properties [152]. However, the multiplicity of soil physicochemical parameters (soil texture, organic matter, nitrogen and phosphorus availability, etc.) made it difficult to accurately determine generalized applicable factors that affect soil microbial communities, and even in many cases, contradictory results are obtained. For example, pH is a key factor influencing the geographic distribution pattern of microbial communities, by regulating microbial community function and thus affecting soil carbon cycling [212]. However, in humid environments, changes in soil moisture are often the dominant factor affecting microbial community structure [112, 213]. Zhang et al., (2020) considered that spatial is more important than temporal in shaping microbial community at large scales and that rapid turnover of soil factors contributes more to temporal variation [214]. Thus, the main factors affecting microbial community distribution are closely related to habitat and soil substrate availability. In this study, we found that there were significant differences in the distribution of microbial community between different land-use types in the reservoir's buffer strips, while the microbial community  $\beta$  diversity in woodland and abandoned cropland was also distributed on distinct sides of coordinate axis at different sites, indicating that there were different mechanisms for the distribution of microbial community at distance scale in these two land-use types, which was consistent with the  $\alpha$  diversity of microbial community observed in these two land-use

types, and it may imply that the grassland soil microbial community had more resilience in water level fluctuation environment. In addition, we found that soil physicochemical properties appeared to be closely related to the specific phylum of the microbial community, with *Actinobacteria* and *Verrucomicrobia* correlating extremely well with four components of soil physicochemical properties that we decomposed, respectively. Notably, *Actinobacteria* was correlated with soil slow turnover chemical properties, while *Verrucomicrobia* was correlated with fast turnover chemical properties, suggesting that the effect of soil physicochemical properties on microbial communities may be through the action of some key microbial taxa [213]. *Actinobacteria* and *Verrucomicrobia* are resistant to water stress, possibly due to their unique physiological traits that allow them to survive in the habitat of desiccation and high moisture fluctuations [215].

The results of the CCA analysis revealed that TC was the main factor that affecting soil microbial communities of different land-use types in the reservoir's buffer strips, which is consistent with the result of Li et al., (2022) in the Three Gorges Reservoir for paddy and dryland [179]. As a primary source for microorganism energy and metabolism, TC play an important environmental filtering role for microbial growth and colonization. Also, riparian buffer strips have been widely demonstrated to have significant carbon loss mechanisms at the distance scale and between land use [131]. Therefore, the accumulation and loss of soil carbon by alternating dry-wet water environment in the reservoir's buffer strips is a key factor limiting the dispersal capacity of the microbial community population. This suggested that strengthening restoration and management practices for soil organic carbon for different land-use types in the reservoir's buffer strips is essential for regulating microbial community structure. It also has important

implications for the management of adopted agroforest ecosystems in the reservoir's buffer strips.

### 6.3.3. Mechanisms of soil microbial community assembly for different land-use types in reservoir's buffer strips

The reservoir's buffer strips are a vulnerable and recovering ecosystem, and its microbial community composition and structure are facing restoration and reconstruction due to frequent water level fluctuation. The resistance and resilience of different land-use types to environmental perturbations will have a profound impact on the recovery process and assembly mechanism of microbial communities [216]. In this study, the NIT index was greater than zero for different land-use types in the reservoir's buffer strips, indicating that the dry-rewet riparian environment promotes microbial community phylogenetically related taxa tend to co-occur more than expected by chance, suggesting that pressure from the environment is the main factor shaping the structure of soil microbial community in the reservoir's buffer strips, which is consistent with the results of numerous studies in riparian zone [106, 217]. We also found that the woodland soil microbial taxa in the reservoir's buffer strips showed a deplected trend compared to the other two land-use types, which indicated that the soil microbial community population of adopted agroforest ecosystems in reservoir buffer strips have a more stable microbial composition. Meanwhile, the neutral community model showed that stochastic processes dominate the grassland soil microbial community construction, which indicated that the adaptation of the grassland microbial community to the environment might be driven by stochastic processes such as dispersal and drift.

Co-occurrence networks can reflect the interactions between microbial communities and taxa in different habitats in terms of structure and function, and are also potential indicators for evaluating the restoration and resilience of microorganisms to environmental disturbances [218]. The complexity of microbial co-occurrence networks is expressed in the topological parameters, such as degree, centralization and clustering coefficient, etc., which depict the size and intrinsic frame of the network [219]. In this study, we found that the abandoned cropland microbial community had a more complex network structure, and the grassland microbial community network tended to be relatively simplified; the grassland had large network parameters such as Diameter and Path Length, all which implied that the interrelationships among the grassland microbial community network seemed to be incompact. While the Centralization Betweenness and Centralization Closeness is also large in the grassland microbial community network suggesting that key microbial taxa in grassland may perform an important role in community stability. We also found higher positive links in abandoned cropland and woodland, suggesting that these two habitats may be more exposed to stress from environmental disturbance, as evidence from the stress gradient hypothesis (SHG) proposed that positive links in the microbial network are more promoted when subjected to external stresses, increasing resource utilization [220, 221]. Meanwhile, ecological models demonstrated that negative links can promote microbial community network stability [222], and the higher negative links in woodland suggested that adopted agroforest ecosystems in the reservoir's buffer strips may have a more stable network structure and greater resilience to the environment than abandoned cropland.

Interestingly, from the results of the neutral community model, we found that

stochastic processes dominate the microbial community assembly mechanism in grassland, yet when decomposing the microbial community assembly mechanism across different habitats, we found that the deterministic process of homogeneous selection occupied quite a significant proportion in grassland. This result was not contradictory, which is mainly due to the disparity in the emphasis of the two approaches on microbial community assembly mechanisms, with the neutral community model proposed by Sloan et al., (2006) highlighting the role of stochastic processes in shaping community structure [223, 224], while the phylogeny-base MNTD highlighting deterministic processes. Actually, this result was more in accord with the complex reality of the reservoir's buffer strips. It was clear that the pressure from the external environment integrally shapes the microbial community of various habitats along the shoreline and determines the direction of the succession of microbial community; Simultaneous, since the resistance and resilience ability of different land-use types to the environment, which limited the ability of microorganism in different habitats to dispersal randomly.

#### 6.4. Conclusion

We investigated the characteristics of microbial community composition and distribution for different land-use types in the reservoir's buffer strips of Chushandian reservoir at fine distance scale and the relationship with soil properties, elucidating the main mechanism driving microbial community assembly processes. The results showed that the microbial community composition and distribution characteristic of abandoned cropland and woodland were quite different at fine distance scales in the reservoir's buffer strips; the alternating dry-rewet shoreline environment promoted the microbial community metabolism-related function in different land-use types, especially in woodland, which implied the adopted agroforest ecosystems in reservoir's buffer strips had more stronger resilience to environmental alternation. Meanwhile, in terms of microbial community assembly mechanism, the stress from environmental alteration was the main pressure that shaped the soil microbial communities of different land-use types in the reservoir's buffer strips, and grassland seemed to have more resilience capacity to disturbance, which was mainly dominated by neutral processes such as stochastic dispersal. Soil microbial community composition and distribution in the reservoir's buffer strips were the results of a combination of deterministic processes from environmental stresses and stochastic processes from the tolerance ability of different habitats to the environment. Among them, TC is the main driving factor affecting microbial community structure.

#### **CHAPTER 7**

### YEILD AND ECONOMIC PRODUCTIVITY FOR ADOPTED AGROFOREST ECOSYSTEM IN RESERVOIR'S BUFFER STRIPS COMPARED TO TRADITIONAL FARMING PRACTICE

7.1. Results

### 7.1.1. The yeild and economic income for adopting agroforest ecosystems in the reservoir's buffer strip

Through investigation, we found that the tea and chestnut species in this area were mainly "Xinyang Maojian" and "689", this is mainly related to the promotion of local agricultural management departments. The planting years of the two species are relatively consistent, especially for tea which has a long history of local cultivation, is one of the top10 famous tea species in China and has high economic values. These two species reached the full fruit stage. Compared to monoculture, the yeild of both species in agroforest ecosystems decreased slightly, with Tea from 215.15 to 184.85 kg/ha, Chestnut from 1499.25 to 852.94 kg/ha, respectively. Production input in agroforest ecosystems is 4997.00 RMB/ha which is lower than Tea monoculture, and higher than Chestnut monoculture. The total economic unit price of agroforest ecosystems for Tea is 55455 RMB/ha, Chestnut 4264.7 RMB/ha, respectively, which were slightly lower than Tea monoculture (56052.75 RMB/ha) and Chestnut monoculture (6282.10 RMB/ha). The net income for agroforest ecosystems is 54722.7 RMB/ha, which is slightly lower than tea monoculture (56052.75 RMB/ha) and greatly higher than Chestnut monoculture

	Adopted Agroforest Ecosystems		Monoculture	
	Tea	Chestnut	Tea	Chestnut
Planting age (year)	20	8	20	10
Species	Xinyang Maojian	689	Xinyang Maojian	689
Yeild (kg/ha)	184.85	852.94	215.15	1499.25
Input (RMB/ha)	4997.00		8492.25	1214.15
Average unit price (RMB/kg)	300	5	300	5
Economic income (RMB/ha)	55455	4264.7	64545	7496.25
Net income (RMB/ha)	54722.7		56052.75	6282.10

Yeild and economica incomes for adopted agroforest ecosystems and monoculture

#### 7.2. Discussion

### 7.2.1. The yeild and economic productivity of adopted agroforest ecosystems in the reservoir's buffer strips compared to monoculture

Chestnut tea intercropping is an important agroforest ecosystem in Xinyang City, Henan Province. Chestnut is a light-favoriting deciduous deep-root tree, tea tree is a shade-tolerant evergreen root shallow small shrub. From the aspect of the biological habits of these two species, the populations of the intercropping system of chestnut and tea are complementary and coordinated in space, which can make better use of space resources. Although, there is partial overlap in time but no major competition; In terms of function, it not only makes the tea garden ecosystem rich and colorful, but also makes the ecological environment conducive to plant growth and improvement of tea quality. Thus, the mixing of chestnut and tea is scientific and reasonable [225]. In this study, we found that the yiled of tea and chestnut for adopted agroforest ecosystems in the reservori buffer strips was decreased compared to monoculture. This differ with previous studies, Liu et al., (2007) proposed that chestnut and tea interplanting can improve the yeild of tea significantly [226]. It indicated that adopted agroforest ecosystems in the reservoir's buffer strips may have certain negative effects on the yeild. But we thought this argument is open to question, notice that the input of adopted agroforest ecosystems in the reservoir's buffer strips is 4997.00 RMB/ha, greatly lower than tea monoculture (8492.25 RMB/ha) which means that the farmer did not take enough investment. We also found that the net income for adopted agroforest ecosystems in the reservoir's buffer strips is 54722.7 RMB/ha, nearly close to the net income of tea monoculture (56052.75 RMB/ha), which indicated that adopted agroforest ecosystems in reservoir's buffer strips can still maintain high economic productivity [227].

#### 7.3. Conclusion

Numerous studies have shown that intercropping of chestnut and tea can effectively improve soil fertility and enzyme activity, promote root growth, improve the microenvironment, and improve tea quality and yeild [225,228-230]. When adopted chestnut and tea intercropping were in the reservoir's buffer strips, it can be seen that the yeild for chestnut and tea all have a certain degree of reduction, which reflected the negative effects of the dynamic moisture environment on crop yeild. While adopted agroforest ecosystems in the reservoir's buffer strips can still maintain relatively higher net income even the farmers took negative passive management measures which indicated that adopted agroforest ecosystems in the reservoir's buffer strips still have great potential for development [231].

This provides evidence for the urgency of further research on the operation model and management of adopted agroforest ecosystems in the reservoir's buffer strips. Addition, the agroforest ecosystem was chestnut and tea intercropping in this study, considering the huge difference in average unit price between these two species, the average unit price of tea is even 60 times than chestnut, exploring species allocation patterns with higher economic value may improve the economic productivity for adopted agroforest ecosystems in reservoir's buffer strips.

#### CONCLUSIONS

Agroforest ecosystems as efficient and synergistic use of resources and eco-friendly sustainable agriculture management measures have been widely emphasized and promoted in Europe, North America, and other developed countries. Due to the unique habitat characteristic of the riparian (reservoir) buffer zone, the trade-off between agriculture production and ecological environment should be fully considered when adopted agroforest ecosystems in this area [232]. However, there are still controversies and theoretical gaps of adopted agroforest ecosystems in the reservoir's buffer strips, which limit its popularization and application [233].

Hence, this study reviewed the current development status and application potential of adopted agroforest ecosystems in the reservoir's buffer strips; Focused on the agroforestry management practice in the newly formed Chushandian reservoir's buffer strips in Huan River catchment of Xinyang City, Henan Province, China, and compared soil physicochemical properties, soil ecology, plant species population, soil aggregates stability, microbial species composition and community ecological processes for adopted agroforest ecosystems in the reservoir's buffer strips to other land-use types, contrasted the yeild and economic productivity for adopted agroforest ecosystems in the reservoir's buffer strips to monoculture, revealed the ecological features and environmental performance of adopted agroforest ecosystems in the reservoir's buffer strips; Clarified the ecological advantages and feasibility for adopted agroforest ecosystems in the reservoir's buffer strips. The main results are synthesized as follows:

1. The dissertation provides solid evidences for the feasibility of adopted agroforest ecosystems in the reservoir's buffer strips; Adopted agroforest ecosystems in the reservoir's buffer strips are effective way to solve the 'human-land conflict' in the reservoir's buffer strips, which can maximize the environmental benefits while safeguarding the agricultural output and has great potential for application in the management and operation of the riparian zone in the future.

2. The dissertation proves that adopting agroforest ecosystem in the reservoir's buffer zone has the advantage of increasing vegetation biodiversity and maintaining high species population; Adopted agroforest ecosystems have the most abundance of plant species (27) than the other land use types, which means adopted agroforest ecosystems in the reservoir's buffer strips can maintain higher vegetation community biodiversity, and support more trees and shrubs growth, improve plant productivity.

3. The dissertation clears adopted agroforest ecosystems in the reservoir's buffer strips can maintain soil vitality and improve the riparian habitat quality; The MBC (239.52 mg/Kg), MBN (27.75 mg/Kg) and DOC (18.38 mg/Kg) content in woodland were higher than the other land use types, which indicated that adopted agroforest ecosystems in the reservoir's buffer strips can maintain higher soil microbial activity. Woodland showed lower NH<sub>4</sub>-N (39.55 mg/kg) content and higher NO<sub>3</sub>-N (3.62 mg/kg) than the other land-use types, which indicated that adopted agroforest ecosystems in the reservoir's buffer strips can inhibit the denitrification process, mitigate greenhouse gas emission. The BD (0.99 g/cm3) and SWC (19.28 %) in woodland were lower than the other land-use types which indicated that adopted agroforest ecosystems in the reservoir's buffer strips can effectively reduce soil water content, accelerate water infiltration.

4. Adopted agroforest ecosystems in the reservoir's buffer strips can maintain higher soil microbial activity, improve soil carbon stock potential. The TC (14.78 g/Kg)

and TN (1.307 g/Kg) content in woodland were higher than the other land-use types which indicated that adopted agroforest ecosystems in the reservoir's buffer strips can improve soil carbon sequestration potential. We also found that the Simpson index, Distance to the watercourse, MBN were the main factors that influenced the soil properties in the reservoir's buffer strips. Further, we evidenced that the land use type influenced soil properties through soil stoichiometric and distance to the watercourse influenced soil properties through plant biodiversity, which provided implications for the key scientific management measures of adopted agroforest ecosystems in the reservoir's buffer strips.

5. The dissertation confirms adopted agroforest ecosystems in the reservoir's buffer strips can maintain stable soil structure, regulate carbon stock change; The soil texture of the agroforest ecosystems buffer was classified as sandy loam; The MWD (W0: 572.00  $\mu$ m, W2: 491.65  $\mu$ m, W20: 539.14  $\mu$ m) in woodland was higher than the other land-use types, which indicated that the adopted agroforest ecosystems in the reservoir's buffer strips can promote aggregates stability. The OCP content (4468.07 g/m2) in W20 was highest than all the other sites, which means the adopted agroforest ecosystems in the reservoir's buffer strips can promote carbon storage capacity.

The silt particle size associated carbon content in grassland and abandoned cropland were significant lower than sand and silt particle size associated carbon, while woodland not shows this trend, which indicated that the adopted agroforest ecosystems in the reservoir's buffer strips can prevent carbon loss. We also found that the silt particle size associated with carbon (TC2) was correlated with most of the soil physicochemcial indicators and was the main factor influencing soil particle size distribution and aggregate

stability in the reservoir's buffer strips, which indicated that the TC2 may play the role of carbon turnover property intermediary. This implies the management carbon cycle when adopted agroforest ecosystems in the reservoir's buffer strips.

6. The dissertation verifies adopted agroforest ecosystems in the reservoir's buffer strips can improve microbial diversity, maintain stable community structure and enhance microbial metabolic activity. The most abundant phylum in agroforest ecosystems reservoir's buffer strips is *Proteobacteria*, *Acidobacteriota*, *Actinobacteria*, and *Chloroflexi*, and the abundance of *Actinobacteriota* and *Methylomirabilota* were significantly greater in woodland than grassland. Adopted agroforest ecosystems in the reservoir's buffer strips has the distinct characteristic for microbial community distribution at fine-scale, which also emphasizes the importance of regionalized management for adopted agroforest ecosystems in the reservoir's buffer strips.

In contrast to agroforest ecosystems, more ASVs were enriched in abandoned cropland and grassland, and the top 5 abundance classified at Phylum level were *Acidobacteriota, Actinobacteriota, Proteobacteria, Chloroflexi,* which indicated that adopted agroforest ecosystems in reservoir's buffer strips has relative stability microbial community composition. The co-occurrence network shows that adopted agroforest ecosystems have low diameter (5.569) and clustering coefficient (5.689), higher negative links (872), which indicates adopted agroforest ecosystems in reservoir's buffer strips can maintain more stability of microbial community structure.

The main process that dominated microbial community structure in agroforest ecosystems was homogeneous selection, followed by dispersal limitation. The microbial community functional abundance of Cell motility, and Signal transduction in agroforest ecosystems were significantly lower and Metabolism of terpenoids, polyketides, Carbohydrate metabolism, Amino acid terpenoids and polyketides were significantly higher than the other land-use types which indicated that adopted agroforest ecosystems in the reservoir's buffer strips have stronger metabolic activity to maintain the functional stability of microbial community. Further, we found TC is the main driving factor affecting microbial community structure in the reservoir's buffer strips.

7. The dissertation demonstrates adopted agroforest ecosystems in the reservoir's buffer strips can maintain stable economic income; The yeild of both species in agroforest ecosystems decreased slightly, Tea from 215.15 to 184.85 kg/ha, Chestnut from 1499.25 to 852.94 kg/ha, respectively, which indicated that the negative effects of dynamic moisture environment on yeild when adopted agroforest ecosystems in the reservoir's buffer strips. The net income for adopted agroforest ecosystems in the reservoir's buffer strips is 54722.7 RMB/ha, nearly close to the net income of tea monoculture (56052.75 RMB/ha), which indicated that adopted agroforest ecosystems in the reservoir's buffer strips can still maintain high economic productivity. Given the extensive management of agroforest ecosystems in the reservoir's buffer strips, we believed that there is still considerable potential for increased yeilds and economic productivity.

8. Finally, the suggestions and matters for attention in management are given; Although adopted agroforest ecosystems in the reservoir's buffer strips have numerous advantages and great potential, while we still need to be cautious. We recommend that farmers should be paid attention to strengthening the management of near-water areas to improve soil quality and maintain land productivity. In addition, the use of phosphorus-containing compounds (fertilizers, pesticides et.,) should be strictly prohibited because and improving the application of biological control measures, as far as possible the use of organic farming.

#### RECOMMENDATIONS

#### For agriculture productor:

1. Although dynamic water level fluctuations will reduce the yeild and productivity of adopted agroforest ecosystems in the reservoir's buffer strips, they can still maintain relatively higher economic incomes. Especially if you are willing to put more effort into scientific management, such as organic farming, it has great potential to exceed the monoculture of economic incomes.

2. Actively exploring species allocation patterns with high economic value may significantly increase your income when adopting agroforest ecosystems in the reservoir's buffer strips. In addition, adopted agroforest ecosystems in the reservoir's buffer strips should pay attention to the implementation of regional management, especially to strengthen the management of near-water areas which can improve the resistance ability to environmental disturbance.

3. Adopted agroforest ecosystems in the reservoir's buffer strips must be paid strict attention to the application of pesticides and fertilizers, especially the use of phosphorus compounds. Please consider biological control measures and organic farming as much as possible and improve the level of management.

#### For the department of environmental management (ecological component):

1. Adopted agroforest ecosystems in the reservoir's buffer strips is an efficient ecological management model. In the case of scientific guidance, it will not cause

damage to the ecological environment but will improve the stability of the shoreline, effectively intercept nitrogen source pollution, and mitigate greenhouse gas emissions.

2. Adopted agroforest ecosystems in the reservoir's buffer strips can prevent soil carbon loss, improve soil carbon sequestration potential, reduce soil moisture condition, increase soil infiltration, enhance microbial activity, enhance microbial resilience to environmental disturbances, and ensure soil health and quality.

3. Adopted agroforest ecosystems in the reservoir's buffer strips can maintain high vegetation biodiversity and shorten the buffer width setting.

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233.Yan, T. Agroforestry system adopts in riparian zones / Materials of the All-Ukrainian scientific conference of students and postgraduates dedicated to the International Student Day (November 14 - 18, 2022). Sumy: SNAU, 2022. P. 68.

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### APPLICATIONS

### Appendix A

#### List of works published on the topic of the dissertation

## LIST OF WORKS PUBLISHED ON THE THEME OF THE DISSERTATION

# Articles in scientific publications included in the list of specialized scientific publications of Ukraine

1.**Yan, T.**, Kremenetska, Ye. O., Wan, S., Hu, Q. & He, S. Ecological functions and environmental benefits of reservoir riparian zone. *Bulletin of Sumy National Agrarian University. The series "Agronomy and Biology"*, Volume 2 (44). 2021. P. 73-82. <u>https://doi.org/10.32845/agrobio.2021.2.10</u> (*The applicant particapated in search literatures, conclustion the results, preparation of article for printing*).

2.**Yan, T.**, Kremenetska, Ye. O., Wan, S., Hu, Q. & He, S. Study of community structure and distribution of mixed forest near Nanwan lake. *Bulletin of Sumy National Agrarian University. The series "Agronomy and Biology"*, Volume 3 (45). 2021: 78–86. <u>https://doi.org/10.32845/agrobio.2021.3.10</u> (*The applicant participated in research, experiment design, analysis of the results and writing the article*).

3.**Yan, T.**, Kremenetska, Ye. O., Wan, S., Hu, Q. & He, S. Soil chemical properties and phytodiversity of riparian forest land near Nanwan lake. *Bulletin of Sumy National Agrarian University. The series "Agronomy and Biology"*, Volume 4 (46). 2021: 97-104. <u>https://doi.org/10.32845/agrobio.2021.4.14</u> (*The applicant participated in research, experiment design, analysis of the results and writing the article*).

## Articles in periodical scientific publications indexed in the Web of Science Core Collection and/or Scopus databases

4.**Yan, T.**; Kremenetska, Y.; Zhang, B.; He, S.; Wang, X.; Yu, Z.; Hu, Q.; Liang, X.; Fu, M.; Wang, Z. The Relationship between soil particle size fractions, associated carbon distribution and physicochemical properties of historical land-use types in newly formed reservoir's buffer strips. *Sustainability*. 2022, 14, 8448.

<u>https://doi.org/10.3390/su14148448</u> (*The applicant participated in research, experiment design, analysis of the results and writing the article*).

5. Yan, T.; Kremenetska, Y.; He, S.; Melnyk, T.; Melnyk, A. The relationship between soil physicochemical properties and vegetation biodiversity in newly formed reservoir's buffer strips. *Applied ecology and environmental research* 2023, 21(4):3153-3175. DOI: <u>http://dx.doi.org/10.15666/aeer/2104\_31533175</u> (*The applicant participated in research, experiment design, analysis of the results and writing the article*).

## **Other publications**

6.**Yan, T.** Agroforestry – an important meaning solve world environment problem // Proceedings of the All-Ukrainian scientific conference of students, dedicated to the International Student Day (November 11-15, 2019). Sumy: SNAU, 2019. P. 424. (The applicant participated in search literature, conclusion the results and preparation the article for printing).

7. Yan, T., Kremenetska, E.O. A review of research progress on forest growth and yeild models / Abstracts of reports of the participants of the international scientific-practical conference "Research of forest and urban ecosystems for sustainable development" (Kyiv, September 22, 2020). Kyiv: NULES, 2020. P. 73-74. (*The applicant participated in search literature, conclusion the results and preparation the article for printing*).

8. Yan, T. Reservoir riparian zone - an important buffer zone of the ecosystem // Proceedings of the All-Ukrainian scientific and practical conference of students and post-graduate students, dedicated to the International Student Day (November 16-20, 2020). Sumy: SNAU, 2020. P. 52. (*The applicant participated in search literature, conclusion the results and preparation the article for printing*).

9. Yan, T., Kremenetska E. The understanding of geochemical process of reservoir riparian zone: the way to water quality / Proceedings of the scientific-practical conference of teachers, graduate students and students of Sumy NAU (April 19-23, 2021). Sumy, 2021. P. 77. (*The applicant participated in search literature, conclusion the results and preparation the article for printing*)

10. **Yan, T.,** Kremenetska E. Soil fertility of reservoir riparian zone / Proceedings of the scientific-practical conference of teachers, graduate students and students of Sumy NAU (April 19-23, 2021). Sumy, 2021. P. 78. (*The applicant participated in search literature, conclusion the results and preparation the article for printing*).

11. Yan, T., Kremenetska, E. O. The restoration and reconstruction of vegetation is

the key to ecological balance of reservoir riparian zone / Proceedings of the III International scientific and practical internet conference "Directions of development of technological systems and logistics in agro-industrial production". Kharkiv: Petro Vasylenko Kharkiv National technical university of agriculture (April 22, 2021). P. 114-116. (*The applicant participated in search literature, conclusion the results and preparation the article for printing*).

12. Yan, T., Kremenetska, E. O. Carbon and nitrogen coupling is the key factor for the stability of riparian ecosystem / Proceedings of the International scientific and practical conference «HONCHARIVSKI CHYTANNYA» dedicated to the 92 th anniversary of Doctor of Agricultural Sciences professor Mykolay Dem'yanovych Honcharov, 25 May 2021. Sumy: SNAU, 2021. P. 219-221. (*The applicant participated in search literature, conclusion the results and preparation the article for printing*).

13.**Yan, T.,** Kremenetska Ye. How does dam construction affect cultivated land? The implication for agricultural production and riparian zone management / International scientific conference «The latest scientific achievements in the modern agro-industrial complex »: conference proceedings (December 28–29, 2021. Lublin, the Republic of Poland). Riga, Latvia: "Baltija Publishing", 2021. P. 32-36. <u>https://doi.org/10.30525/978-9934-26-184-8-8</u> (*The applicant participated in search literature, conclusion the results and preparation the article for printing*)

14.Yan, T., Kremenetska Ye. Vegetation community characteristic and construction mechanism of reservoir riparian zone / Proceedings of the International Scientific and Practical Conference "HONCHARIVSKI CHYTANNYA", dedicated to the 93 rd anniversary of Doctor of Agricultural Science professor Honcharov Mykola

Dem'yanovych, 25 May 2022. Sumy: SNAU, 2022. P. 195-197. (*The applicant participated in search literature, conclusion the results and preparation the article for printing*).

15.Yan, T. Agroforestry system adopts in riparian zones / Materials of the All-Ukrainian scientific conference of students and postgraduates dedicated to the International Student Day (November 14 - 18, 2022). Sumy: SNAU, 2022. P. 68.

(The applicant participated in search literature, conclusion the results and preparation the article for printing).

16.**Yan, T.**, Kremenetska Ye. The key role of microbial communities in riparian buffer strips / Proceedings of the International Scientific and Practical Conference "HONCHARIVSKI CHYTANNYA", dedicated to the 94 rd anniversary of Doctor of Agricultural Science professor Honcharov Mykola Dem'yanovych, 25 May 2023. Sumy: SNAU, 2023. P. 230-232. (*The applicant participated in search literature, conclusion the results and preparation the article for printing*).

## Appendix B

Latin Name	Genera	Family	Importance Value			
			Cropland	Grassland	Woodland	
Erigeron annuus	Compositae	Erigeron L.	0.35	0.28	0.12	
Conyza canadensis	Compositae	Leucosoma	0.20	0.10	-	
Imperata cylindrica	Gramineae	Imperata	0.36	0.28	0.34	
Veronica persica Poir	Scrophulariaceae	Veronica L.	0.18	0.20	0.09	
Ceratium glomeratum Thuill	Caryophyllaceae Juss	Cerastium	0.42	0.56	-	
Pteris ensiformis Burm	Pteridaceae	Pteris	-	-	0.18	
Callitriche palustris L.	Callitrichaceae	Callitriche	-	-	0.06	
Senecio scandens BuchHam. ex D. Don	Compositae	Senecio	0.02	-	0.04	
Rosa sp	Rosaceae	Rose	-	-	0.02	
Carex L	Cyperacea	Carex	0.01	0.28	-	
Geranium wilfordii Maxim	Geraniaceae	Erodium	-	-	0.07	
Duchesnea indica (Andr.) Focke	Rosaceae	Duchesnea	-	-	0.06	
Kalimeris indica	Compositae	Kalimeris	-	-	0.04	
Spiraea salicifolia L	Rosaceae	Spiraea	-	-	0.03	
Diaphasiastrum veitchii	Lycopodiaceae	Lycopodiella	-	-	0.09	
Castanea mollissima Bl.	Fagaceae	Castanea Mill	-	-	0.29	
Phyllostachys glauca McClure	Gramineae	Phyllostachys Sieb.	-	-	0.22	
Ophiopogon bodinieri Levl	Liliaceae	Ophipogon Ker-Gawl.	-	-	0.10	

The vegetation species composition in Chushandian reservoir's buffer strips

:Rubus parvifolius L	Rosaceae	Rubus	-	0.06	0.08
Cocculus orbiculatus (L.) DC	Menispermaceae	Cocculus	-	-	0.02
Rhododendron simsii Planch.	Ericaceae	Rhododendron	-	-	0.01
Boehmeria nivea (L.) Gaudich	Urticaceae	Boehmeria	-	-	0.04
Lamium amplexicaule L	Labiatae	Lamium	-	0.05	-
Al, ternanthera Philoxeroides(Mart.)Gris eb	Amaranthaceae Juss	Alternanthera Forsk.	-	0.33	-
Eleusine indica	Gramineae	Eleusine	-	0.24	-
Capsella bursa-pastoris	Brassicaceae	Capsella	-	0.01	0.03
Lagopsis supina (Stephan					
ex Willd.) IkonnGal. ex	Labiatae	Marrubium L.	-	0.03	-
Knorring					
Vicia sepium Linn	Leguminosae	Vicia	-	0.03	0.04
Youngia japonica	Compositae	Youngia	-	0.01	0.08
Salvia plebeia R. Br.	Labiatae	Salvia	-	0.02	0.01
Artemisia argyi	Compositae	Sagebrush	-	0.23	-
Hydrocotyle sibthorpioides Lam	Mbelliferae	Hydrocotyle L.	0.15	-	-
Lonicera japonica	Caprifoliaceae	Lonicera L.	0.26	-	-
Cirsium japonicum Fisch. ex DC.	Compositae	Cirsium	0.02	-	-
Campanula medium L	Campanulaceae	Campanula	0.04	-	-
Semiaguilegia adoxoides (DC.) Makino	Ranunculaceae	Semiaquilegia	0.15	-	-
Mentha haplocalyx Briq	Labiatae	Mentba L.	0.07	-	-
Reineckia carnea	Liliaceae	Reineckia	0.01	-	-
Hippochaete	EquisetumL	Equisetum	0.12	-	-

hiemale(L.)Boerner					
Ophiopogon japonicus	Liliaceae	Ophipogon	0.13	-	-
(Linn. f.) Ker-Gawl		Ker-Gawl.			
Mazus japonicus (Thunb.)	Scrophulariaceae	Mazus	-	0.14	-
O. Kuntze					
Sonchus asper	Compositae	Sonchus L.	-	0.02	-
Calystegia hederacea	Convolvulaceae	Calystegia	-	0.03	-
Wall	Juss				
Cirsium setosum	Compositae	Cirsium	-	0.03	-
Digitaria sanguinalis (L. )	Gramineae			0.07	
Scop.		Digitaria Scop.	-	0.07	-
Carduuscrispus L.	Compositae	Carduus	-	0.02	-
Stellaria media (L.) Cyr.	Caryophyllaceae Juss	Stellaria	-	-	0.08
Vicia hirsuta	Leguminosae	Vicia	-	-	0.06
Poa annua L	Gramineae	Poa	-	-	0.03
Morus alba L.	Moraceae	Morus	-	-	0.02
Torilis scabra (Thunb.)	Mbelliferae	Torilis Adans.	-	-	0.06
DC.					
Lysimachia christinae	Primulaceae	Lysimachia	-	-	0.03
Hance					
Glycine soja Sieb. et		Glycine L.	-	-	0.02
Zucc.	Leguminosae				
Euphorbia esula Linn.	Euphorbiaceae	Euphorbia L.	0.14		
Juncus effusus	Juncaceae	Juncus L.	0.21		
Lindernia procumbens (K	Scrophulariaceae	Lindernia	0.02		
rock.) Philcox					
Euphorbiahelioscopia L.	Euphorbiaceae	Euphorbia L.	0.11		