THE INFLUENCE OF RICE-DUCK FARM SOIL ON THE NITROGEN CONTENT OF DIFFERENT VARIETIES OF JAPONICA RICE

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ABSTRACT

Nitrogen fertilizers are widely applied to increase rice yields, but excessive fertilization poses an environmental risk. It has been shown that rice-duck farming can be more efficient in terms of N use by improving rice growth. Several pathways have been proposed for how ducks may improve rice growth in paddy fields. The aim of this study was to investigate the isolated effect of rice-duck farm soil on rice, namely the N content in different plant organs and whether it differs among rice varieties. In a 116-day greenhouse pot experiment, six different Japonica rice varieties (JinU99, Jinyuan98, Jindao18, Jinyuan89, Jinhei1 and Jindao201) were grown in fertilized duck and no-duck soil after which the N content in their organs and the numbers of surviving and grain-producing plants were compared. The straw and leaf N concentrations were positively influenced by rice-duck farm soil while in the roots, this effect was recorded in only two rice varieties. Grain N content differed among varieties, but was not significantly influenced by soil type. Overall, N concentrations in straw and leaf, and roots of some rice varieties, but not in grain grown on duck soil were higher than that in those grown in soils not influenced by rice-duck farming. This study for the first time demonstrates that rice-duck farm soil alone can influence rice growth, namely an increase in the N content of certain rice plant organs.

Keywords: co-culture; nitrogen; rice plant; soil

Introduction

Rice is the most important cereal crop for more than 3 billion people in the world and for about 60% of the Chinese population (Xiong et al. 2013). China is the largest producer of rice (Frolking et al. 2002). Between 1977 to 2005, the total annual grain production in China increased by 71% to 484 million tons (Ju et al. 2009). While the human population is increasing in China and across the world, cultivable land resources are limited. Addressing food security requires novel strategies to increase crop production, including sustainable fertilizer use strategies.

Nitrogen (N) fertilizer is used to enhance rice production (Zhang et al. 2012). However, nitrogen use efficiency is relatively low in rice fields because of N losses via ammonia volatilization, denitrification, surface runoff and leaching of the soil in floodwater systems (Vlek and Byrnes 1986). These losses represent a substantial environmental threat because of the emissions of ammonia and greenhouse gases and groundwater pollution (Bijay-Sing and Craswell 2021). Avoiding these losses is an important goal for the development of environmentally focused rice cropping strategies.

Previous studies have shown that rice-duck farming can lead to lower N losses and higher N use efficiency and improved rice growth than conventional rice farming (Yu et al. 2009; Gao et al. 2019). The rice-duck co-culture has more than a 400-year-old history in China and has been widely adopted in many other Asian countries such as Japan, South Korea, Malaysia and Philippines because of its economic, environmental and ecological benefits (Zheng et al. 2016). The presence of ducks in paddy fields promotes rice production and quality (Suh 2014; Teng et al. 2016; Li et al. 2017). Ducks control weeds and pests (Liu et al. 2004; Quan et al. 2005) and promote soil fertility via their droppings (Teng et al. 2016). Moreover, ducks provide mechanical effects including plowing, muddying and mechanical stimulation of rice by engaging in their activities including walking, swimming, eating, grooming, paddling and rubbing (Luo and Gliessman 2016). Duck activities not only stimulate rice growth, but can also increase its lodging resistance (Zhang et al. 2013). Understanding the mechanisms by which ducks influence rice plants in paddy fields is necessary to continue improving rice-duck farming systems towards higher N use efficiency. For example, rice-duck farming can induce higher N content in certain rice organs (Ebissa et al. 2018). Several direct and indirect pathways are proposed for why ducks influence rice growth, based on field observations of rice-duck farming and conventional rice cultivation. In such conditions, the different pathways may act together and their isolated effects cannot be tested. No study has to our knowledge focused on isolating the effect of rice-duck farming soil on rice growth.

The main objective of the current study was to investigate the effect of rice-duck farm soil on rice seedling survival and rice N content and whether it differs among dif-

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Table 1 Times and amounts of fertilizer applied at the Ninghe experimental farm.

	Basic fertilizer	Transplanting	Tiller fertilizer		
Date	25-April	10-May	17-May	26-May	17-June
Fertilizer	$(NH_4)_2HPO_4$:	Rice seedlings	$(NH_4)_2SO_4$:	$(NH_4)_2SO_4$:	$(NH_2)_2CO$:
Rate (kg ha^{-1})	112.44		149.25	5.97	1.12
Rate (kg N ha^{-1})	23.84		31.63	1.27	5.22

$(NH_4)_2HPO_4$: ammonium diphosphate; $(NH_4)_2SO_4$: ammonium sulphate; $(NH_2)_2CO$: urea

ferent Japonica rice varieties. We hypothesized that rice plants grown in duck soil will have a higher N content.

Materials and Methods

Study site and experimental design

The study was carried out using soil and rice seedlings collected at the Ninghe experimental farm (39°18′–39°50′ N, 117°08′– 117°56′ E), located in North China. At the experimental farm, two areas were studied: duck and no-duck fields (each 3 × 7 m). The amounts of fertilizer applied at the farm are shown in Table 1. In the duck field, twenty ducks were released at the vegetative stage in 2017. Soil samples for soil characterization were collected from each field using an auger in October 2017 as described in Ebissa et al. (2018). For the purpose of the present study, only the 0–20 cm layer was sampled, yielding three samples from each of four plots in both the duck and no-duck fields (n = 12 per treatment). In April 2018, seeds of six Japonica rice varieties (JinU99, Jinyuan98, Jindao18, Jinyuan89, Jinhei1 and Jindao201) were sown at the farm in an area not influenced by ducks close to the two fields. In June 2018, rice seedlings were collected together with soil from the surface layers of each of the two fields.

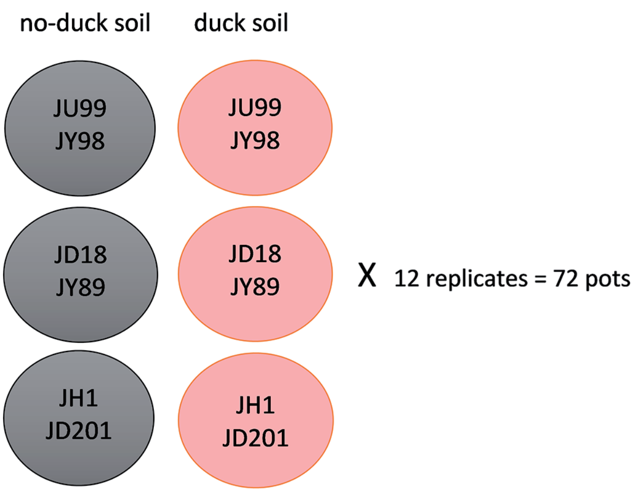

Fig. 1 Experimental design of the greenhouse pot experiment. Pots with either duck soil or no-duck soil were each planted with two seedlings of 6 varieties using the following pattern. JU99: Jin U99, JY98: Jinyuan 98, JD18: Jindao 18, JY89: Jinyuan 89, JH1: Jinhei no. 1, JD201: Jindao 201.

The rice seedlings and the duck and no-duck soil were then brought to a greenhouse for an indoor pot experiment. Treatments were arranged in a total of 72 pots and included 2 types of soil (duck vs. no-duck) and 3 pairs of rice varieties (JU99+JY98, JD18+JY89, JH1+JD201) in 12 replicates (Fig. 1). Each pot (∅ 22 cm, height 22 cm) contained approximately 5 kg of soil and two seedlings, one of each variety. Fertilizers were applied to each pot: 0.75 g (urea), 0.25 g (calcium superphosphate) and 0.5 g (potassium sulfate) on June 29, 2018 (day 1 of the experiment). The same amounts of fertilizer were applied on day 34, amounting to a total N application of 321 N kg ha^{-1}. The rice plants were watered 3 to 5 days a week until maturity and harvested on October 22, 2018 (day 116 of the experiment).

The harvested rice plants were sorted into grain, straw, leaf and root. The roots were washed to avoid any contamination with soil. The rice plant samples were oven-dried for three days at 75 °C and ground to fine powder. The soil samples were air-dried, ground and sieved to pass through a 0.15-mm mesh. The following soil properties were evaluated: pH, total N, NO_3^-, NH_4^+, soil organic matter, total P and particle size distribution, as described in Ebissa et al. (2018) (Table 2).

Data analysis

All data were analyzed using R software. A two-way ANOVA was conducted to examine the interaction between soil types and rice variety on N concentrations in each plant. A paired t-test was used to compare survival

Table 2 Physicochemical properties (±SD, n = 12) of soil at 0–20 cm depth in the experimental fields (2017). SOM – soil organic matter.

Property	Unit	Duck soil	No-duck soil	t-test
pH		7.42 ± 0.09	7.48 ± 0.04	ns
Total N	(g kg^{-1})	1.04 ± 0.19	0.93 ± 0.26	ns
NH_4^+–N	(mg kg^{-1})	3.92 ± 4.46	2.23 ± 0.94	ns
NO_3^-–N	(mg kg^{-1})	25.98 ± 15.48	35.66 ± 14.96	ns
Total P	(g kg^{-1})	0.89 ± 0.16	0.91 ± 0.11	ns
SOM	(g kg^{-1})	22.75	19.60	
Clay	(%)	32	35	
Silt	(%)	63	56	
Sand	(%)	6	9	

ns: non-significant

and grain production of varieties. An unpaired t-test was used to compare the soil properties of the duck and no-duck fields.

Results and Discussion

The main objectives of this study were to investigate the effect of rice-duck farm soil on rice seedling survival and rice N content and whether it differs for different rice varieties.

Number of surviving and grain-producing plants

Out of the 12 rice seedlings of each variety at the beginning of the experiment, between 9 and 12 survived to the end of the experiment (Fig. 2). Highest survival was recorded for varieties JD18, JY89 and JU99 and the lowest for JY98 and JD201, without any major effect of soil. Not all the seedlings produced grain at the end of experiment, with the number ranging between 4 and 12 for different varieties and soil types (Fig. 2). The number of grain-producing plants was higher in duck soil than no-duck soil for half of the varieties (JY89, JH1, JD201) while the opposite was true for the other half (JU99, JY98, and JD18). However, neither survival nor grain production of the varieties were significantly influenced by soil type (paired t-test, $p > 0.05$). JD18 produced the highest number of grain-producing plants in duck soil while in no-duck soil this was also true for JU99. To our knowledge, there is no study that compares the suitability of different rice varieties for rice-duck co-culture and our preliminary data indicate that some varieties might be more suitable than others. Future studies should however focus also on the potential differences in biomass and grain yield of the varieties and include also monocropping treatments to exclude the potential effects of competition.

N concentrations in rice plants

In line with our hypothesis, the N concentrations in rice plants were influenced either by soil type, the variety

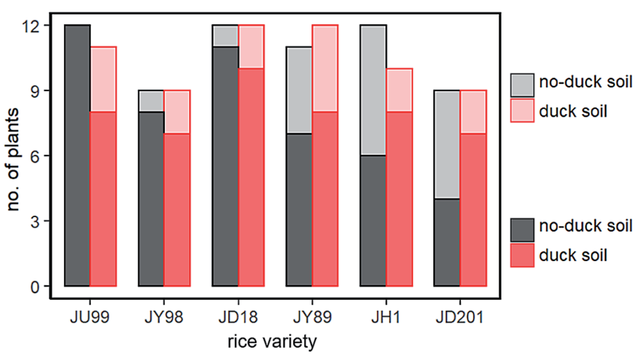

Fig. 2 Number of plants of each variety that survived (light-coloured) and produced grain (dark-coloured) at the end of the 116-day pot experiment. Twelve seedlings were planted at the beginning of the experiment.

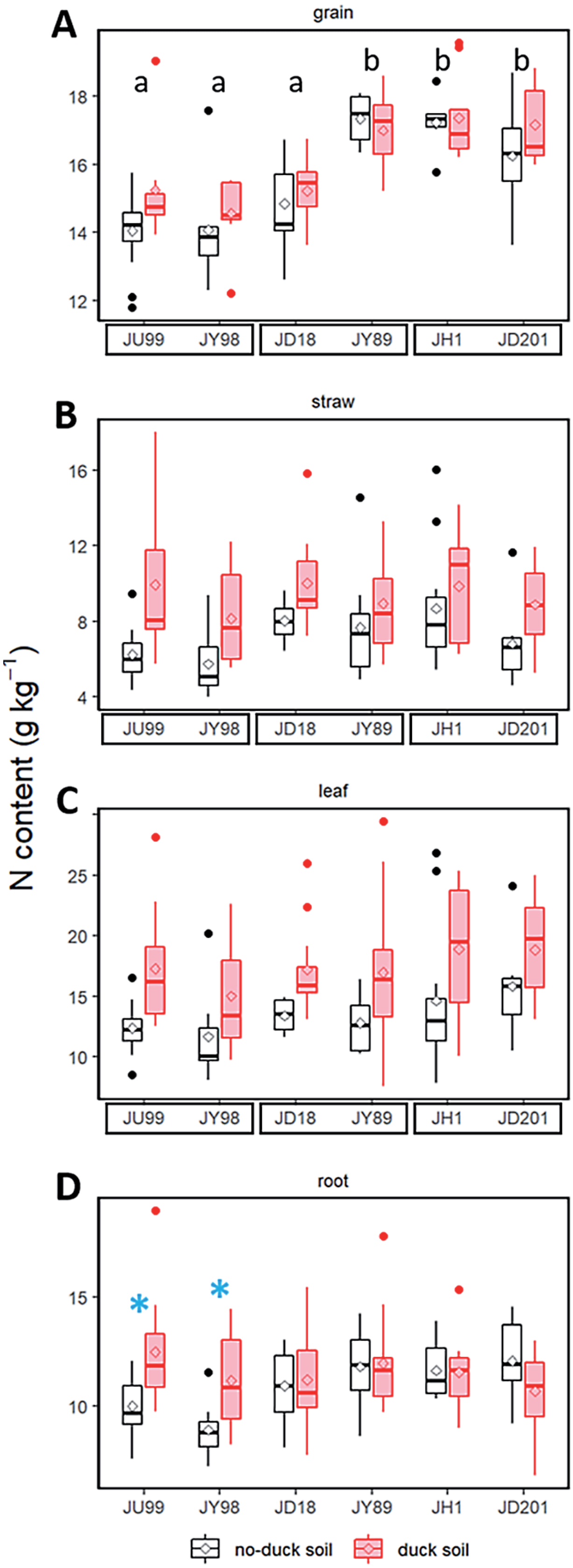

Fig. 3 Nitrogen content of grain (A), straw (B), leaves (C) and roots (D) of six varieties of Japonica rice grown in soil from a rice-duck field (duck soil) and a field without ducks (no-duck soil). Only significant main or interaction effects of two-way ANOVA are listed, see Table 1 for details. *, ** and *** denote significance at 0.05, 0.01 and 0.001 probability levels, respectively. Different letters and asterisks indicate significant differences (Tukey post-hoc test, $p < 0.05$).

Table 3 The effects of types of soil and variety of rice and their interaction on the N contents of grain, straw, leaves and roots as tested by two-way ANOVA.

Variable	df	N content (g kg^{-1})							
		Grain		Leaves		Straw		Roots	
		F	P	F	P	F	P	F	P
Soil types (ST)	1	3.206	0.0770	25.555	**<0.0001**	21.876	**<0.0001**	3.615	0.0598
Rice variety (RV)	5	18.293	**<0.0001**	1.938	0.0940	2.154	0.0640	1.958	0.0902
ST × RV	5	0.739	0.5960	0.127	0.9860	0.735	0.5980	2.963	**0.0150**

*, **, and *** denote significance at 0.05, 0.01, and 0.001 probability levels, respectively. Different letters and asterisks indicate significant differences (Tukey post-hoc test, $p < 0.05$).

or their interaction depending on the part of the plant (Fig. 3, Table 3). Concentrations in grain did not differ between duck soil and no-duck soil, but were different for the varieties being higher in Jinyuan 89, Jinhei 1 and Jindao 201 than in Jin U99, Jinyuan 98 and Jindao 18 (Fig. 3A). In contrast, straw and leaf N concentrations were higher for plants grown in duck soil (Fig. 3B, C), which is in accordance with our hypothesis. Neither straw nor leaf N concentrations were associated with rice variety. Finally, also the concentrations in roots were higher for plants grown in duck soil, but only in varieties JY99 and JY98 (Fig. 3D). Similarly, to straw and leaf, variety did not affect N concentrations in roots.

The variability in N content was higher in leaf and straw than in grain and root. Not all plants produced grain at the end of the experiment, which might be explained by the plants being in different phases of development. It is reported that the N content of leaves can vary at different stages of the development of rice, ranging between 4.9% to 50 g kg^{-1}, with lowest values at booting and highest at the vegetative stage (Wang et al. 2014; Gholisadeh et al. 2017).

That duck soil can increase the N content of rice plants is in accordance with the results of the preceding field experiment of Ebissa et al. (2018). To our knowledge, no other study has so far compared the N content of rice plants produced by rice-duck and conventional farming, but other related parameters have been studied. Nitrogen is an important component of proteins and N content of leaves is strongly correlated with chlorophyll content (Singh et al. 2002; Wang et al. 2014; Yang et al. 2014), which in turn is related to grain yield (Gholisadeh et al. 2017). In line with our findings, Teng et al. (2016) report higher contents of soluble protein and chlorophyll in duck treatments than in no-duck treatments at certain times during the growing season. Also, Li et al. (2019) report increases in leaf soluble protein content. This corresponds with many other previous studies that have shown the positive effect of rice-duck farming on rice growth and production compared to conventional fields (Liu et al. 2004; Saleh and Seyyed 2015). These effects include increased grain yield (Hossain et al. 2004; Saleh and Seyyed 2015), plant height (Hossain et al. 2004; Saleh and Seyyed 2015), grain number per panicle (Ahmad et al. 2004; Saleh and Seyyed 2015), one thousand grain weight (Karbalaie 2004), number of tillers per hill (Mohammadi et al. 2013) and harvest index (Saleh and Seyyed 2015).

Since we found increased N content in certain rice organs on duck soil, future studies should aim to also quantify N uptake to explore if this corresponds also to higher N use efficiency. In general, the N application rate in paddy fields ranges between 50 and 500 kg ha^{-1} (Che et al. 2015). In our experiment, 321 kg N ha^{-1} was applied to the pots and 62 kg N ha^{-1} was applied in the field before collecting the seedlings (total 383 kg N ha^{-1}) to simulate conventional fertilization rates. Further studies should also explore the potential for fertilizer reduction on rice-duck farm soil as a next step towards environmentally focused rice cropping.

Soil in the duck and no-duck fields

In order to understand the mechanisms resulting in the higher N content of rice grown in duck soil, soils collected from duck and no-duck fields were analyzed. We expected the duck soil to have higher content of organic matter and nitrogen and thus act as soil enriched with organic fertilizer. However, we found no significant differences ($p > 0.05$) between the duck and no-duck field in any of the investigated soil properties related to soil fertility, namely soil pH, total N, NH_4^+ and NO_3^-, and total P (Table 2, note that difference in SOM could not be tested). This contrasts with the findings of Yang et al. (2004) who report that soil from rice-duck co-culture was higher in soil organic matter, total N, available N as well as available P and K. Teng et al. (2016) report the effect of rice-duck farming on the availability of nutrients in the soil (namely NH_4^+, alkali hydrolysable N and available P) throughout the growing season and conclude that differences only occur in September and are not present in October (when the soil was sampled in present study). This could explain the discrepancy at least in the highly dynamic available N pools.

We expected to find higher total and available N pools in the duck soil since it is estimated that the total excreted faeces per duck can reach 10 kg, which contains 47 g

N, 70 g P and 31 g K (Xiong and Zhu 2003). On a daily basis, fresh droppings of a duck weigh an average 0.14 kg and contain 7.1 g N kg^{-1} (Long et al. 2013). However, a study in which duck faeces alone or in combination with mineral fertilizer were added to soil showed no positive effects on rice growth, yield or protein content (Isobe et al. 2005), suggesting that the nutrient content alone may not be the most important factor determining the fertility of rice-duck farm soil. Increase in soil enzyme activity or soil physical characteristics such as lower bulk density and higher aggregation in duck-influenced soil (Yang et al. 2004) may be equally or more important, but were not measured in the present study. Overall, we can conclude that in our study we did not find any differences in the soil parameters of duck and no-duck fields. Future studies should consider wide range of soil properties (including different soil organic matter fractions, enzyme activity or soil physical characteristics) and a targeted analysis of the soil at the start and at the end of the pot experiment.

Direct and indirect effects of ducks on rice growth

This study is the first to confirm that rice-duck farming soil alone can positively affect rice growth, namely the nitrogen content of the rice plants. Previous studies were conducted only in the field where the ducks could have influenced rice growth by various direct and indirect pathways. For example, water and soil disturbance caused by ducks walking in rice fields may inhibit weed germination. This can result in up to 99% weed reduction after four years of rice-duck cultivation. Ducks are also effective at reducing pests (Quan et al. 2005) while at the same time are not harmful or even beneficial in terms of arthropod diversity (Qin et al. 2011) or nematode abundance (Teng et al. 2016). Stimulating effects of duck activities on rice plants can cause some changes in shape, height, stalk thickness, effective tillering and other growth characteristics of rice plants (Takao 2001; Shen 2003; Zhang et al. 2007, 2011; Wang et al. 2008; Huang et al. 2012; Zhang et al. 2012). Last but not least, ducks improve soil properties (Yang et al. 2004). Apart from the effect of duck droppings on soil chemistry, the movement and feeding activity of ducks in rice plots causes variations in soil distribution, thus resulting in improving soil physical properties, which subsequently improve the root systems of rice plants (Furuno 1996).

Differences in the responses of the rice varieties

This study is also the first to compare the responses of different varieties of Japonica rice to rice-duck farming soil; however, the differences were small. The compared Japonica rice varieties did differ in grain N content but this was irrespective of soil type. Similarly, some differences were found also in the survival of rice plants, again irrespective of the soil. Only the root N content response to duck soil differed among the varieties, but a monocropping study would be needed to assess if these differences affect yield, biomass production and N uptake.

Conclusion

Rice-duck farming can improve rice growth via several pathways. The objective of this study was to investigate the effect of rice-duck farm soil on rice N content and its variability among rice varieties. We found that N concentrations in certain organs of the rice varieties grown in duck soil were higher compared to those grown in no-duck soil. Namely, N concentrations in straw, leaves and roots of some rice varieties were higher when grown in duck soil. Neither survival or grain production of the varieties were significantly influenced by soil type. Our study is the first to confirm that duck-influenced soil can promote rice growth, namely its nitrogen content. We give several recommendations on further research towards the application of rice-duck farming for sustainable rice production. Further experiments should investigate grain yield, biomass, and wider range of soil properties in response to different fertilization rates to further assess the effect of duck soil on rice growth and nitrogen use efficiency.

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