

Effects of the white stork friendly farming method on plant community diversity in paddy fields in the Tajima region of Japan

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Abstract In the paddy field ecosystem, plants provide food and shelter for animals and support biodiversity. However, the diversity of plant communities in paddy fields is declining. In the Tajima region of Hyogo Prefecture, Japan, farmers have been practicing the “White Stork Friendly Farming (WSFF) method” for eco-friendly rice production since 2003. This method aims to conserve biodiversity within the rice production system and to support the reintroduction of the Oriental White Stork (*Ciconia boyciana*). This study assessed the effect of the WSFF method on plant community diversity in paddy fields. Plant surveys were conducted in June, July, and August/September to identify plant species within sample plots in the paddy fields and levees managed under the WSFF method and conventional rice production method, followed by an assessment of plant species diversity using the Shannon–Weiner index. The sample plots were classified using two-way indicator species analysis, and the relationship between the sample plots and indicator species was identified through detrended correspondence analysis. The WSFF method contributed to an increase in plant community diversity within paddy fields, indicating a positive impact on the conservation of biodiversity in these fields through rice production.

Key words Farming method, Paddy field, Plant community diversity

Introduction

Rice paddy fields provide substitute habitats for plants and animals living in wetland ecosystems in Japan (Hidaka 1998; Natuhara 2013; Ohtani et al. 2013). In the paddy field ecosystem, plants play a crucial role by providing food and shelter for animals, thereby supporting biodiversity (Okubo and Maenaka 1995). Unfortunately, plant community diversity in these paddy fields has been declining (Kudoh 2017).

Aimed at restoring biodiversity in paddy fields, a subset of farmers in the Tajima region of Hyogo Prefecture practices the White Stork Friendly Farming (WSFF) method for eco-friendly rice production. This method was established in 2003 by the Hyogo prefectural government to support the reintroduction of the Oriental White Stork (*Ciconia boyciana*). The areas of rice paddies cultivated using this method reached 445 hectares in 2022 (Toyooka City 2022). The WSFF method comprises rice production and water management practices aimed at conserving biodiversity in paddies. These practices include nonuse or reduced-use of agrochemicals, flooding paddy fields in winter and early spring, deep-water management (i.e., keeping irrigated water at a depth of 8 cm for 40 days after seedling transplant), and mid-summer drainage conducted a couple of weeks later than the conventional rice farming method (Hyogo Prefecture 2019; Nishimura and Ezaki 2019).

Studies have examined the impacts of land improvement and differences in farming practices on plant species composition in paddy fields (Noguchi 1992; Okubo and Maenaka 1995; Yamaguchi et al. 1998; Ito et al. 1999; Matsumura 2002). Naito et al. (2020) reported that the WSFF method is associated with an increased number of plant species occurring in paddy fields. However, research on the effects of the WSFF method on plant diversity remains scarce. In this context, we hypothesize that the WSFF method increases plant species diversity in rice paddies. Our aim is to assess its effect in both levees and paddy fields.

Materials and Methods

1. Study site

We conducted this study in Toyooka, Tajima region of Hyogo Prefecture, Japan, focusing on three study sites: Mie, Nitta, and Gonosho (Fig. 1). At each site, 20 paddy field areas were selected: 10 under a conventional method (Conv.) and 10 under the WSFF method. This resulted in a total of 60 paddy field areas sampled. We identified these farming methods

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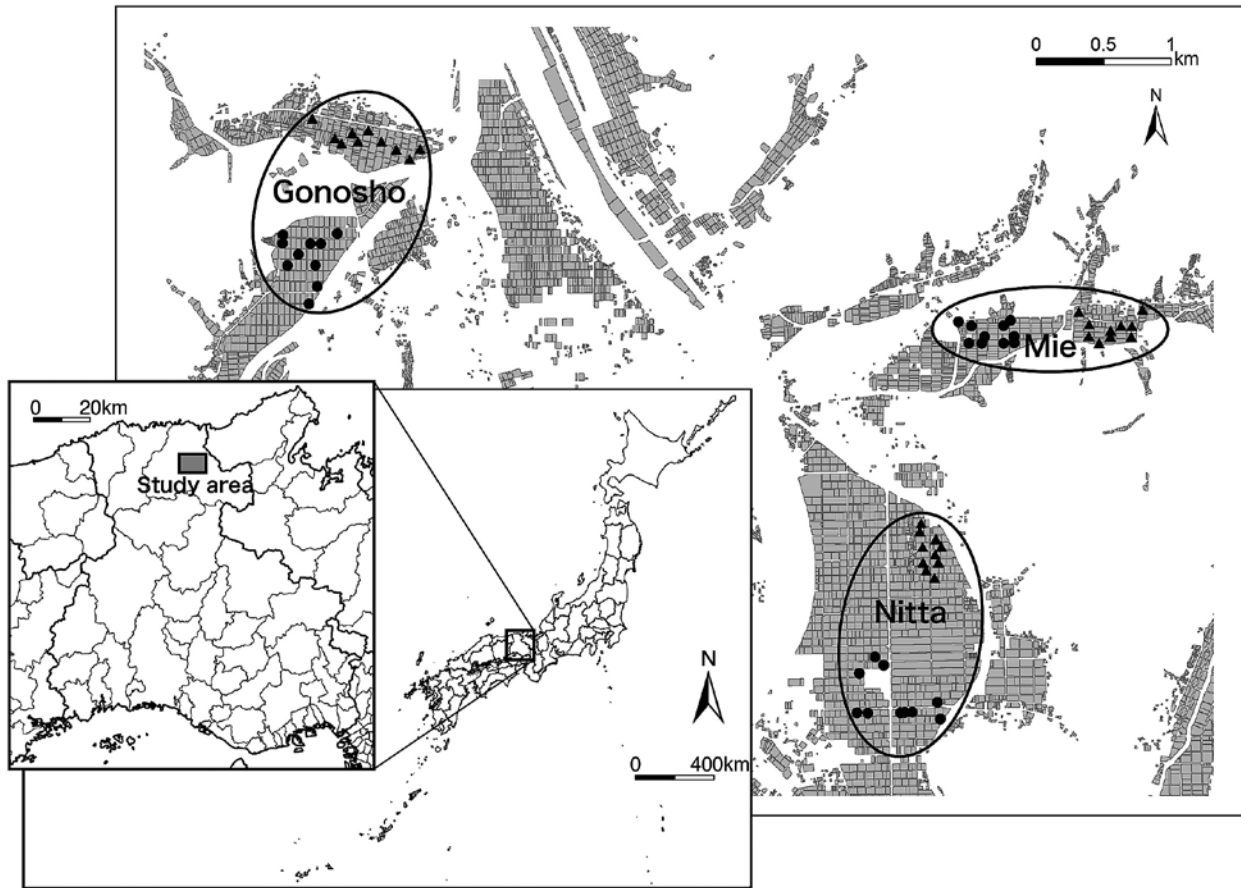


Fig. 1. Administrative area maps showing study site locations (lower left corner) and three rice paddy districts (Gonosho, Mie, and Nitta), each encompassing 20 rice paddies (10 conventional paddy fields and 10 eco-friendly paddy fields, represented by circles and triangles, respectively). These maps were created by processing digital national land information from the Ministry of Land, Infrastructure, Transport and Tourism, Japan (Ministry of Land, Infrastructure, Transport and Tourism, Japan, 2022).

based on the information provided by land managers.

2. Methods

We randomly selected three sides out of four levees from each of a rectangular-shaped paddy field and established a sample plot of 1 m² quadrat both in the levee and within the paddy field in each of the selected sides of the paddy field, which resulted in three sample plots both in the levee and within the paddy field for each paddy field. In total, 180 sample plots were established in the levees and 180 plots within the paddy fields, respectively. The sample plots within the paddy fields were placed within the paddy fields at around 1 m inside from the levee where rice seedlings (*Oryza sativa*) were planted. The vascular plant species in the sample plots were identified and their coverage in percentage was estimated through visual assessment. Plant surveys were conducted within paddy fields in (1) early June (just after rice seedling transplant), (2) late July, and (3) between late August and mid-

September in 2016 (prior to rice harvesting). We undertook plant surveys in the levees (1) between late June and early July, and (2) between late September and early October. The sample size for the second levee survey was 174, as adverse weather conditions and the subsequent mowing of study plots impeded the survey of two paddy fields using the conventional method in the Nitta area. The sample plots were established randomly for each plant survey. The levees in the study sites were mowed once in a month and a half, or four times during a cropping season.

The Shannon–Weiner index (H') of the sample plots for assessing plant species diversity was calculated using Equation 1.

$$H' = -\sum p_i \times \ln(p_i) \quad (1)$$

where p_i is the proportion of the total cover of the i th species. We calculated the average H' value for each paddy field by averaging the H' values from three plots within that field, representing the plant community diversity of the

Table 1. Number of native and alien species found in paddy fields under the different farming methods.

Habitat	Order of survey	Conv.		WSFF		Total	
		Native	Alien	Native	Alien	Native	Alien
Within the paddy	1st	8	2	7	1	10	2
	2nd	24	2	21	3	30	3
	3rd	35	4	28	3	37	4
In the levee	1st	70	23	61	11	79	23
	2nd	88	15	85	10	87	16

Conv., Conventional method; White Stork Friendly Farming method.

respective paddy field. For each study site, the Mann–Whitney U test was used to determine differences in the values of H' between the paddy fields of the conventional method and those of the WSFF method. H' was calculated using R version 4.2.1 (R Development Core Team 2022) and the U test by SPSS (IBM SPSS Statistics 28.0.0.0). A two-way indicator species analysis (TWINSpan) (Hill 1979) was used to classify the sample plots into groups with similar species abundance and composition. The sample plots and plant species were subjected to detrended correspondence analysis (DCA) for ordination (Hill and Gauch 1980). We conducted both analyses using PCORD version 7 (McCune and Grace 2002).

Results

In the first plant survey within the paddy fields in June, 12 non-*O. sativa* species were identified. The total numbers of native and alien species found in the paddy fields managed under the conventional method were 8 and 2, respectively, and those under the WSFF method were 7 and 1, respectively (Table 1). Among them, species with a high frequency of occurrence (i.e., the number of times they occurred in 180 sample plots) included *Azolla cristata x filiculoides* (15%),

Lemna aoukikusa (8.3%), *Monochoria vaginalis* (7.2%), and *Spirodela polyrhiza* (6.7%). The first plant survey within the paddy fields revealed that 67 plots (74.4%) using the conventional method and 51 plots (56.7%) using the WSFF method contained no species other than *O. sativa*. In addition, the average number of species, including *O. sativa*, per plot was 1.36 (SD = 0.74) for the conventional method and 1.67

Table 2. Average number of species occurring in paddy fields under the different farming methods.

Habitat	Order of survey	Gotosho		Nitta		Mie		Total	
		Conv.	WSFF	Conv.	WSFF	Conv.	WSFF	Conv.	WSFF
Within the paddy	1st	1.13 (±0.43)	1.13 (±0.35)	1.30 (±0.54)	1.63 (±0.85)	1.63 (±1.03)	2.23 (±0.97)	1.36 (±0.74)	1.67 (±0.89)
	2nd	2.83 (±1.64)	3.77 (±1.14)	3.57 (±1.36)	4.97 (±1.45)	3.73 (±1.93)	3.67 (±1.56)	3.38 (±1.69)	4.13 (±1.50)
	3rd	5.80 (±1.85)	6.17 (±1.86)	5.60 (±2.08)	8.50 (±1.80)	5.57 (±2.40)	6.60 (±1.69)	5.66 (±2.10)	7.09 (±2.04)
In the levee	1st	10.00 (±3.13)	10.87 (±2.57)	10.87 (±2.49)	11.97 (±2.55)	11.07 (±4.29)	13.80 (±2.80)	10.64 (±3.38)	12.21 (±2.88)
	2nd	12.03 (±3.36)	13.67 (±3.40)	12.33 (±2.57) ¹⁾	12.70 (±3.00)	10.47 (±2.40)	13.33 (±3.07)	11.56 (±2.91) ²⁾	13.23 (±3.15)

Conv., Conventional method; White Stork Friendly Farming method.

Standard deviations are presented in parentheses.

¹⁾The sample size of the plots of each method in each study site was 30 except for that of the conventional method in Nitta study site for the second survey in the levee (n = 24).

²⁾The sample size of the plots of each method in total was 90 except for that of the conventional method for the second survey in the levee (n = 84).

Table 3. Average cumulative total plant cover per plot in the paddy fields under the different farming methods.

Habitat	Order of survey	Gonosho			Nitta			Mie			Total		
		Conv.	WSFF	Conv.	WSFF	Conv.	WSFF	Conv.	WSFF	Conv.	WSFF	Conv.	WSFF
Within the paddy	1st	0.01 (±0.04)	0.13 (±0.03)	0.13 (±0.04)	0.13 (±0.03)	7.41 (±21.70)	0.89 (±2.01)	2.49 (±12.88)	0.33 (±1.22)				
	2nd	1.44 (±2.16)	5.44 (±6.26)	1.44 (±2.16)	5.44 (±6.26)	1.44 (±3.66)	26.65 (±32.40)	1.32 (±2.55)	20.04 (±24.91)				
	3rd	1.90 (±2.30)	14.96 (±11.50)	1.90 (±2.30)	14.96 (±11.50)	1.86 (±3.37)	21.03 (±23.31)	2.15 (±2.80)	21.05 (±18.66)				
In the levee	1st	78.81 (±31.06)	60.03 (±18.15)	78.81 (±31.06)	60.03 (±18.15)	84.73 (±28.41)	103.16 (±12.22)	80.35 (±28.30)	82.70 (±25.37)				
	2nd	96.29 (±23.37)	128.21 (±28.67)	96.29 (±23.37) ¹⁾	128.21 (±28.67)	98.07 (±15.65)	81.24 (±20.30)	102.93 (±22.08) ²⁾	108.64 (±29.70)				

Conv., Conventional method; WSFF, White Stork Friendly Farming method.

Standard deviations are presented in parentheses.

¹⁾The sample size of the plots of each method in each study site was 30 except for those of the conventional method in Nitta study site for the second survey in the levee (n = 24).

²⁾The sample size of the plots of each method in total was 90 except for that of the conventional method for the second survey in the levee (n = 84).

(SD = 0.89) for the WSFF method (Table 2).

The plant cover of species other than *O. sativa* was small. The average cumulative total plant cover per plot (i.e., the average value of the summation of coverage of species,

excluding *O. sativa*, in the plot) was 2.49% (SD = 12.88) for the conventional method and 0.33% (SD = 1.22) for the WSFF method (Table 3).

Within plots using the conventional method, *A. cristata* x *filiculoides* and *Echinochloa crus-galli* had covers exceeding 10%. In contrast, only *A. cristata* x *filiculoides* had cover exceeding 10% within the plots using the WSFF method.

The second survey in July identified 33 non-*O. sativa* species within paddy fields. The total numbers of native and alien species found in the paddy fields managed under the conventional method were 24 and 2, respectively, and those under the WSFF method were 21 and 3, respectively. Species with a high frequency of occurrence included *M. vaginalis* (43.3%), *Sagittaria trifolia* (32.8%), *Ludwigia epilobioides* (22.2%), and *Lindernia procumbens* (22.2%). The average number of species per plot was 3.38 (SD = 1.69) for the conventional method and 4.13 (SD = 1.50) for the WSFF method. The average cumulative total plant cover per plot was 1.32% (SD = 2.55) for the conventional method and 20.04% (SD = 24.91) for the WSFF method. Only *A. cristata* x *filiculoides* had cover exceeding 10% within the plots employing the conventional method. However, *M. vaginalis*, *S. trifolia*, and *Lindernia dubia* var. *dubia* had cover exceeding 10% in the plots using the WSFF method.

The third survey conducted in August/September uncovered 41 non-*O. sativa* species within paddy fields. The total numbers of native and alien species found in the paddy fields managed under the conventional method were 35 and 4, respectively, and those under the WSFF method were 28 and 3, respectively. Species with a high frequency of occurrence included *L. procumbens* (68.9%), *M. vaginalis* (43.9%), *Persicaria hydropiper* (37.2%), and *L. epilobioides* (36.1%). The average number of species per plot was 5.66 (SD = 2.10) for the conventional method and 7.09 (SD = 2.04) for the WSFF method. The average cumulative total plant cover per plot was 2.15% (SD = 2.80) for the conventional method and 21.05% (SD = 18.66) for the WSFF method. Only *A. cristata* x *filiculoides* and *E. crus-galli* had cover exceeding 10% within the plots using the conventional method. Conversely, *M. vaginalis*, *S. trifolia*, *L. dubia* var. *dubia*, *L. procumbens*, *Paspalum distichum*, *Eleocharis kuroguwai*, and *Ceratopteris gaudichaudii* var. *vulgaris* had cover exceeding 10% in the plots utilizing the WSFF method.

The first plant survey in the levees conducted in June/July

Table 4. Shannon–Wiener diversity indices under the different farming methods.

Habitat	Order of survey	Gonosho		Nitta		Mie		Total	
		Conv.	WSFF	Conv.	WSFF	Conv.	WSFF	Conv.	WSFF
Within the paddy	1st	0.01 (±0.00)	0.01 (±0.00)	0.03 (±0.02)	0.11 (±0.04)	0.19 (±0.10)	0.20 (±0.06)	0.07 (±0.04)	0.11 (±0.03)
	2nd	0.10 (±0.02)	0.31 (±0.06) *	0.09 (±0.02)	0.74 (±0.10) *	0.10 (±0.04)	0.61 (±0.15) *	0.10 (±0.01)	0.55 (±0.07)
	3rd	0.14 (±0.03)	0.65 (±0.08) *	0.18 (±0.03)	0.94 (±0.09) *	0.13 (±0.03)	0.60 (±0.12) *	0.15 (±0.02)	0.73 (±0.06)
In the levee	1st	1.63 (±0.14)	1.84 (±0.11)	1.68 (±0.06)	1.79 (±0.06)	1.77 (±0.17)	1.88 (±0.09)	1.69 (±0.07)	1.84 (±0.05)
	2nd	1.66 (±0.12)	2.03 (±0.11)	1.72 (±0.12) ¹⁾	1.80 (±0.07)	1.45 (±0.11)	1.99 (±0.09) *	1.60 (±0.07) ²⁾	1.94 (±0.06)

Conv., Conventional method; White Stork Friendly Farming method.

Standard deviations are presented in parentheses.

¹⁾The sample size of the plots of each method in each study site was 10 except for those of the conventional method in Nitta study site for the second survey in the levee (n = 8).

²⁾The sample size of the plots of each method in total was 30 except for that of the conventional method for the second survey in the levee (n = 28).

*P < 0.05 by Mann–Whitney U test.

uncovered 102 species in total. The total numbers of native and alien species found in the paddy fields managed under the conventional method were 70 and 23, respectively, and those under the WSFF method were 61 and 11, respectively. The species with a high frequency of occurrence included *Hydrocotyle ramiflora* (85%), *Equisetum arvense* (72.2%), *Trifolium repens* (68.9%), and *Setaria viridis* (51.1%). The average number of species per plot was 10.64 (SD = 3.38) for

the conventional method and 12.21 (SD = 2.88) for the WSFF method. The average cumulative total plant cover per plot was 80.35% (SD = 28.30) for the conventional method and 82.70% (SD = 25.37) for the WSFF method.

The second survey in the levees in September/October identified 103 species. The total numbers of native and alien species found in the paddy fields managed under the conventional method were 88 and 15, respectively, and those under the WSFF method were 85 and 10, respectively. The species with a high frequency of occurrence included *Digitaria ciliaris* (83.9%), *E. arvense* (70.1%), *Kyllinga brevifolia* var. *leiolepis* (67.8%), and *H. ramiflora* (78.2%). The average number of species per plot was 11.56 (SD = 2.91) for the conventional method and 13.23 (SD = 3.15) for the WSFF method. The average cumulative total plant cover per plot was 102.93% (SD = 22.08) for the conventional method and 108.64% (SD = 29.70) for the WSFF method.

For the first survey within the paddy field, the values of H' for the plant community in the paddy fields managed under the conventional method in Gonosho, Nitta, and Mie were 0.01, 0.03, and 0.19, respectively, whereas those in the paddy fields managed under the WSFF method were 0.01, 0.11, and 0.20, respectively (Table 4).

The values of H' for the plant community for the second survey within the paddy fields in the paddy fields managed under the conventional method in Gonosho, Nitta, and Mie were 0.10, 0.09, and 0.10, respectively, whereas those in the paddy fields managed under the WSFF method were 0.31, 0.74, and 0.61, respectively. The values of H' for the plant community for the third survey within the paddy fields managed under the conventional method in Gonosho, Nitta, and Mie were 0.14, 0.18, and 0.13, respectively, whereas those in the paddy fields managed under the WSFF method were 0.65, 0.94, and 0.60, respectively. For the first survey in the levees, the values of H' for the plant community in the paddy fields managed under the conventional method in Gonosho, Nitta, and Mie were 1.63, 1.68, and 1.77, respectively, whereas those in the paddy fields managed under the WSFF method were 1.84, 1.79, and 1.88, respectively. The values of H' for the plant community in the second survey in the levees in the paddy fields managed under the conventional method in Gonosho, Nitta, and Mie were 1.66, 1.72, and 1.45, respectively, whereas those in the paddy fields managed under the WSFF method were 2.03, 1.80, and 1.99, respectively.

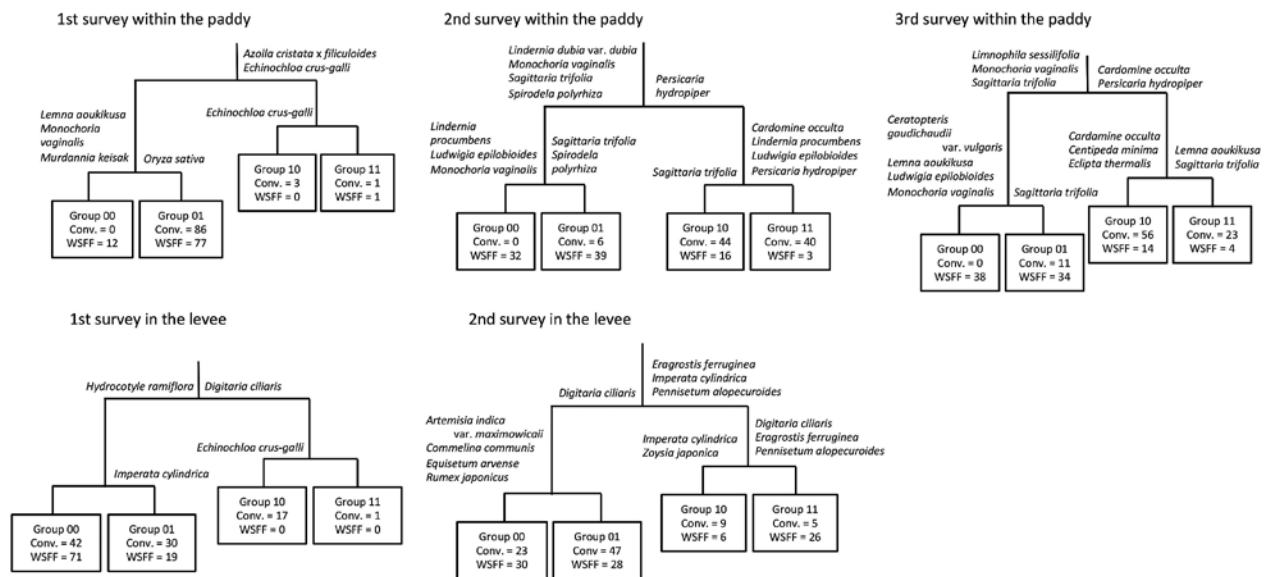


Fig. 2. Classification dendrograms of plant community types identified by TWINSpan. The number attached to Group is the code name of the group. Indicator species for the negative (0) or positive (1) group and number of conventional and/or WSFF plots in each division are also shown.

The first survey within the paddy fields revealed no significant difference in H' values between the paddy fields managed under conventional and those managed under the WSFF methods. However, the second and third plant surveys within the paddy fields revealed that the values of H' for the plant community in the paddy fields managed under WSFF method significantly exceeded those in the paddy fields managed under the conventional method in all three study sites. The first survey in the levees revealed no significant difference in H' values between the paddy fields managed under the two methods. The H' values for the plant community in the second levee survey in the paddy fields managed under the WSFF method markedly exceeded those in the paddy fields managed under the conventional method, but this difference was observed only at the Mie study site.

The TWINSpan results classified the sample plots of each plant survey into four groups at the second level of division (Fig. 2).

For the first survey within the paddy fields, the TWINSpan classified the plots into 2 groups (Group 0 and Group 1) at the first split with *A. cristata x filiculoides* and *E. crus-galli* as positive indicators. Group 0 was further divided into 2 groups (Group 00 and Group 01) with *L. aoukikusa*, *M. vaginalis*, and *Murdannia keisak* as negative indicators and with *O. sativa* as a positive indicator. Group 01 was the largest group, with 163 plots where almost only *O. sativa* occurred. Group 00 was a small group that only included WSFF plots. The other two

groups (Group 10 and Group 11) were very small and included only 3 and 2 plots.

For the second survey within the paddy fields, the TWINSpan classified the plots into 2 groups (Group 0 and Group 1) at the first split with *L. dubia var. dubia*, *M. vaginalis*, *S. trifolia*, and *S. polyrhiza* as negative indicators and with *P. hydrogiper* as a positive indicator. Group 0 mainly included WSFF plots and was further divided into 2 groups (Group 00 and Group 01) with *L. procumbens*, *L. epilobioides*, and *M. vaginalis* as negative indicators and *S. trifolia* and *S. polyrhiza* as positive indicators. The Group 00 included only WSFF plots. Group 1 mainly included conventional plots and was further divided into 2 groups (Group 10 and Group 11) with *S. trifolia* as a negative indicator and *Cardamine occulta*, *L. procumbens*, *L. epilobioides* and *P. hydrogiper* as positive indicators.

For the third survey within the paddy fields, the TWINSpan classified the plots into 2 groups (Group 0 and Group 1) at the first split, with *Limnophila sessiliflora*, *M. vaginalis*, and *S. trifolia* as negative indicators and *C. occulta* and *P. hydrogiper* as positive indicators. Group 0 mainly included WSFF plots and was further divided into 2 groups (Group 00 and Group 01), with *C. gaudichaudii var. vulgaris*, *L. aoukikusa*, *L. epilobioides*, and *M. vaginalis* as negative indicators and *S. trifolia* as a positive indicator. Group 00 included only WSFF plots and Group 01 included more WSFF plots than conventional plots. Group 1 was further divided into 2 groups

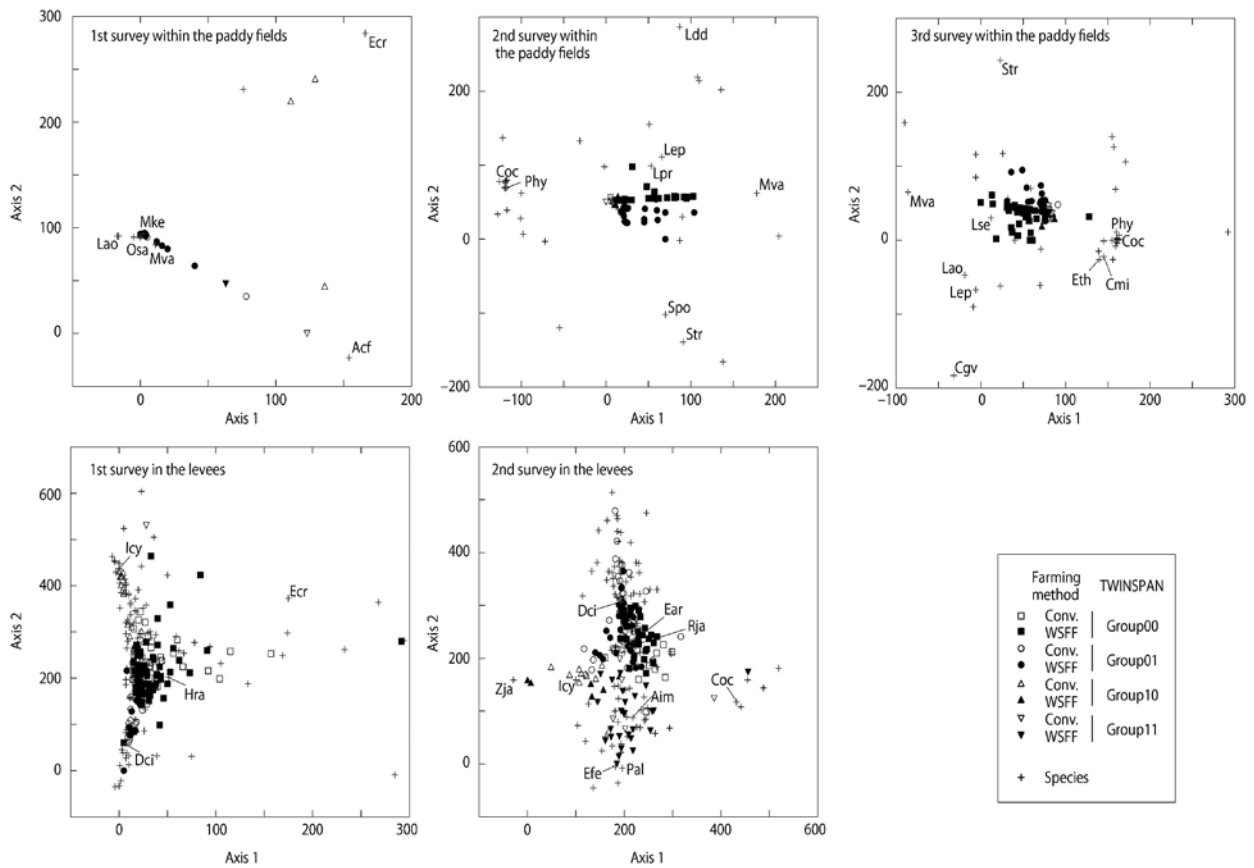


Fig. 3. Diagram of sample plots in DCA ordination. The community types identified by TWINSpan, as well as farming methods, were related to those identified by the DCA using symbols. Indicator species identified by TWINSpan are indicated by abbreviations as follows: Acf: *Azolla cristata* x *filiculoides*, Ecr: *Echinochloa crus-galli*, Lao: *Lemna aoukikusa*, Mva: *Monochoria vaginalis*, Mke: *Murdannia keisak*, Osa: *Oryza sativa*, Ldd: *Lindernia dubia* var. *dubia*, Str: *Sagittaria trifolia*, Spo: *Spirodela polyrhiza*, Phy: *Persicaria hydropiper*, Lpr: *Lindernia procumbens*, Lep: *Ludwigia epilobioides*, Coc: *Cardamine occulta*, Lse: *Limnophila sessiliflora*, Cgv: *Ceratopteris gaudichaudii* var. *vulgaris*, Cmi: *Centipeda minima*, Eth: *Eclipta thermalis*, Hra: *Hydrocotyle ramiflora*, Dci: *Digitaria ciliaris*, Icy: *Imperata cylindrica*, Efe: *Eragrostis ferruginea*, Pal: *Pennisetum alopecuroides*, Aim: *Artemisia indica* var. *maximowiczii*, Cco: *Commelina communis*, Ear: *Equisetum arvense*, Rja: *Rumex japonicus*, Zja: *Zoysia japonica*.

(Group 10 and Group 11), with *C. occulta*, *Centipeda minima*, and *Eclipta thermalis* as negative indicators and *L. aoukikusa* and *S. trifolia* as positive indicators. Both Group 10 and Group 11 mainly included conventional plots.

For the first survey within the levee, the TWINSpan classified the plots into 2 groups (Group 0 and Group 1) at the first split, with *H. ramiflora* as a negative indicator and *D. ciliaris* as a positive indicator. Group 0 was further divided into 2 groups (Group 00 and Group 01), with *Imperata cylindrica* as a negative indicator. Group 1 only included conventional plots and was further divided into 2 groups (Group 10 and Group 11), with *E. crus-galli* as a negative indicator.

For the second survey within the levee, the TWINSpan classified the plots into 2 groups (Group 0 and Group 1) at the first split, with *D. ciliaris* as a negative indicator and *Eragrostis ferruginea*, *I. cylindrica*, and *Pennisetum*

alopecuroides as positive indicators. Group 0 was further divided into 2 groups (Group 00 and Group 01), with *Artemisia indica* var. *maximowiczii*, *Commelina communis*, *Equisetum arvense*, and *Rumex japonicus* as negative indicators. Group 1 only included conventional plots and was further divided into 2 groups (Group 10 and Group 11), with *I. cylindrica* and *Zoysia japonica* as negative indicators and *D. ciliaris*, *E. ferruginea*, and *P. alopecuroides* as positive indicators. Group 11 mainly included WSFF plots, but the other groups did not have clear features in relation to farming methods.

Figure 3 shows the DCA ordination diagram for each plant survey. In the first survey within the paddy fields, the WSFF method had the most plots clustered around the diagram origin. The eigenvalues of the first and second axes were 0.73 and 0.38, respectively. However, in the second and third surveys, the WSFF method showed a widespread distribution

of plots along both axis-1 and axis-2 compared to those of the conventional method. While the eigenvalues of the first and second axes of the second survey were 0.30 and 0.11, respectively, they were in the same order, 0.21 and 0.12, in the third survey. The DCA ordination diagrams of the second and third surveys within the paddy fields also indicated that the plots of the WSFF method were associated with *M. vaginalis* and the plots of the conventional method with *C. occulta*. *M. vaginalis* and *C. occulta* appeared opposite sides along axis-1 on the DCA ordination diagram, and WSFF plots of Group 00 were relatively associated with *M. vaginalis* in both surveys. The diagram of the third survey showed that the plots of the WSFF method mainly including Group 00 were associated with *L. sessiliflora*. *S. trifolia* was associated with the plots of Group 01 on the DCA ordination diagrams of the second and third surveys within the paddy fields. Group 01 mainly included the plots of the WSFF method and some from the conventional method.

The DCA ordination diagrams of the first survey in levees showed that the plots of the WSFF and conventional methods appeared mixed together. *E. ferruginea* and *P. alopecuroides* were associated with the plots of Group 11 on the diagram of the second survey in levees. Group 11 mainly included the plots of the WSFF method and some from the conventional method.

Discussion

The results of plant surveys within the paddy fields did not show significant difference in the total number of species as well as in the number of alien species between the paddy fields managed under the conventional method and those managed under the WSFF method. The results of plant surveys in the levees found that the total number of species as well as the number of alien species were larger in the paddy fields managed under the conventional method than in those managed under the WSFF method. These results suggest that practicing the WSFF method does not increase the number of alien species in the paddy fields.

The first plant survey within the paddy fields found that most plots of both the conventional and WSFF methods had no species other than *O. sativa*. The cover of the non-*O. sativa* species, which were identified in the plots, was small. This resulted in no significant difference in plant community

diversity between the two methods.

Comparing the H' values from the second and third surveys within the paddy fields, our results indicated that plant species exhibited greater diversity within the paddy fields using the WSFF method compared to those using the conventional method. The results of TWINSpan for the second survey within the paddy fields showed that a group of plots, i.e., Group 00, included only the plots of WSFF method. Those plots in the group were relatively widely dispersed along the axis-1 in the DCA ordination diagram. These results suggested that the WSFF method increased plant community diversity. The reduced or no application of herbicides within WSFF paddy fields may be associated with high plant community diversity.

The DCA ordination diagram and TWINSpan results also suggested that the WSFF method was associated with *M. vaginalis* occurrence. These results corroborate those of Naito and Sagawa (2014). The WSFF method aims to suppress weed growth by flooding paddy fields in winter as an alternative to the use of herbicides. However, previous research suggested that winter-time flooding and deep-water management alone were ineffective in controlling *M. vaginalis* (Inaba 1999). Kaneko and Nakamura (2009) also reported that the volume of *M. vaginalis* in paddy fields practicing winter-time flooding markedly exceeded that of paddy fields using the conventional method.

The findings of the present study also revealed a higher occurrence of *L. sessiliflora* in paddy fields using the WSFF method compared to those using the conventional method. Previous research has reported that deep-water management increased *L. sessiliflora* (Tanaka and Kanou 2002). Therefore, the WSFF method may also increase *L. sessiliflora* occurrence owing to deep-water management and reduced or no herbicide application.

Our results also demonstrated that *C. occulta* was more associated with the conventional rice cultivation method. *C. occulta* did not appear either in the paddy fields of the conventional method or in those of the WSFF method for the first survey within the paddies. However, the species was identified in the 15 plots of the conventional method, whereas it was not found in the plots of the WSFF method during the second survey. We also found *C. occulta* in 32 plots of the conventional method and in only one plot of the WSFF method for the third survey. The DCA diagram indicated the

association between the species and conventional method. Previous research has reported that winter-time flooding suppressed *C. occulta* germination (Arihara and Koyama 2002). Therefore, the WSFF method could control *C. occulta* occurrence by winter-time flooding.

Regarding the first plant survey in the levees, the comparison of the diversity indices (H') between the two methods did not yield a significant difference. In the second levee survey, the comparison of indices (H') indicated that the plant species in the levees using the WSFF method exhibited higher diversity, but this difference was significant only at the Mie study site. A previous study reported a positive effect of the WSFF method on species richness and accumulated coverage (Naito et al. 2020). In this study, the WSFF method consistently exhibited higher species richness and H' . However, the differences were sometimes too small to identify significance. These results suggest that the WSFF method may have a modest impact on increasing plant species diversity in paddy field levees, although in some cases, the effect may not be significant. This could be attributed to the absence of herbicide use for weed control in both the conventional and WSFF method paddy field levees.

Conclusion

This study assessed the effect of the WSFF method on plant species diversity as well as on plant community diversity in the paddy fields. The association between the WSFF method and plant species found in the study sites were also explored. Practicing the WSFF method contributed to increasing plant species diversity as well as plant community diversity within paddy fields. This method reduced herbicide application for weed control through deep-water management and winter-time flooding. Identifying additional practices within the WSFF method could enhance its effectiveness in conserving plant community diversity in rice paddies.

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Additional notes

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