Supporting Information

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SI Text

S1. Study Area and Rice–Fish Coculture System. The study was conducted in the area of a "globally important agricultural heritage system" (GIAHS) as designated by the United Nations Development Program and the Global Environment Facility in 2005. The GIAS site is located in southeastern Zhejiang Province (120° 26′–121°41′E, 27°25′–28°57′N), China (Fig. S1*A*). The landscape of the area around the site is diverse and includes coastline, hills, and mountains. The region has a subtropical monsoon climate with a mean annual air temperature of 17–18 °C and a mean annual precipitation of 1,400–1,450 mm. The major cropping system in this area is one-season rice with one crop per year. Of the 45,000 ha in rice production in the five counties of this region, ~10,000 ha are used for rice–fish coculture.

The rice–fish coculture system (Fig. S1*C*) in this GIAHS area was started by local farmers >1,200 y ago. The fish is an indigenous, red, soft-scaled, common species of carp (Oujiang color common carp, *Cypinius carpia color* var.). This fish species, which has high genetic diversity, originated in streams and subsequently evolved in rice fields. Local people consider this fish not as a variety of common carp but as a distinct fish, which they call "Qingtian field fish". The rice varieties in this system change over time. In the last decade, high-yielding hybrid varieties have been planted most often in the region. In this rice–fish coculture system, the fish remain in the rice field all year, except for the few days when rice is transplanted in May and harvested in October. During these days, the fish are moved to a small ditch in a corner of the field.

The GIAHS pilot site (Longxian village, 120°18′E, 27°59′N) (Fig. S1*B*) where we conducted the experiments is within a small watershed with a total area of 300 ha and with 30 ha of rice–fish coculture. The soil has a pH of 6.0, 32.92 $g \cdot kg^{-1}$ organic matter, 2.79 $g \cdot kg^{-1}$ total nitrogen (N), and 23.22 $mg \cdot kg^{-1}$ extractable phosphorus (P).

S2. Observation of Fish Activity and the Removal of Rice Planthoppers by Fish Activity. In 2007, fish activities and air temperature were monitored. We established a quadrat (2 m^{-2}) in each plot of rice–fish coculture (RF) and fish monoculture (FM) (quadrats *A* and *C* in Fig. S5); the quadrat in RF included 16 rice hills. We installed a video recording system near each quadrat and recorded the quadrats from early morning 5:00 AM to evening 6:00 PM (the period when fish are active) on three consecutive sunny days for one block (one replicate, Fig. S5). The total observation time for the whole experiment was 12 d (3 d for each block). We calculated the frequency at which fish visited the quadrat (appearing in the quadrat) in every hour by viewing the tapes on a computer with the Motic Images Advanced 3.2 system.

At the same time that fish activity was monitored, a sampling quadrat including four rice hills was set up in each RF (quadrat *B* in Fig. S5) and RM (quadrat *D* in Fig. S5) plot to record how rice planthoppers were removed by fish. In RF, the video recording system that was documenting the number of times fish visited the quadrat also documented the number of time that fish hit rice stems with their heads. The frequency at which fish hit rice plants was calculated as number of hits per hill per hour by using recordings of quadrat *B*.

Rape seed oil was placed on the surface of the water in each quadrat *B* and *D* to capture the rice planthoppers that fell from rice plants (regardless of what caused them to fall). Like quadrat *A*, quadrat *B*, *C*, and *D* were fenced with floating bamboo rods, which do not affect fish activity but prevent oil on the water surface from diffusing outside of the quadrat (Fig. S5). The planthoppers that were stuck in the oil were collected every hour from 5:00 AM to 6:00 PM and immediately fixed in a mixed liquid with formaldehyde, acetate, and alcohol. Planthoppers were quantified in this manner on three consecutive sunny days for each plot. The fixed planthoppers were transported to the laboratory and counted with a microscope. These data were collected when the rice was at the tillering stage and the average air temperature was 32.6 °C; there were 117 ± 13 and 91 ± 8 planthoppers per hill on plants in RM and RF, respectively.

We found that rice planthoppers fell onto the water surface in RM because of "nonfish" effects (probably wind), and we assumed that these nonfish effects were the same in RM and RF. To identify the numbers of rice planthoppers removed from plants by fish and to estimate removal rate of rice planthoppers, we performed the following calculations:

i) The numbers of rice planthoppers removed from rice plants by fish (numbers per quadrat) = (the numbers of rice planthoppers collected from the water surface in RF quadrats (quadrat *B*, Fig. S5)) − ((the numbers of rice planthoppers collected from the water surface in RM quadrats (quadrat *D*, Fig. S5)).

ii) Removal rate of rice planthoppers by fish (%) = (removal numbers of rice planthoppers by fish per quadrat)/(the total numbers of rice planthoppers on plants at the time of observation per quadrat).

For RF, the total number of rice planthoppers on rice plants in a quadrat at the time of observation was 364 ± 33 (91.4 ± 8.4 per hill × 4 hills per quadrat).

S3. Measurement of Microclimate. At 12:00 PM–2:00 PM of each day from July 29 to August 18, 2007, temperature of surface water and solar radiation intensity under the rice plant canopy were monitored in each plot with five thermometers and three light meter quantum sensors (LI-250; LI-COR). The weather was sunny, and this was the hottest time of year in the area.

S4. Measurement of Ammonia N in Water and Total N in Soil. In each experimental year, we sampled field water from each plot of RF and FM in July, August, and September. After alkaline persulfate digestion (1), ammonia N in these water samples was measured with a flow injection analyzer (Lachat Quickchem Systems). Immediately after the harvest in each year, samples of surface soils (0–20 cm depth) were collected at five random positions in each plot. The soil samples were air dried, ground, and passed through a 0.147-mm sieve. Soil total N was determined by the Kjeldahl method (2).

S5. Analysis of the N Balance. The balance of N output and input within each system (RM, RF, or FM) was calculated by subtracting total N output (N contained in harvested plants and fish) from total input (N contained in fertilizers and fish feed). A positive value would indicate that some portion of the input N was not used by rice and fish but remained in the field plot (as N in the soil or water) or had moved into the surrounding environment (as N in the atmosphere or in drainage water). A negative value would indicate that rice or fish obtained N from sources other than fertilizer or fish feed (from N already in the soil or added to the soil by N fixation or rain deposition).

S6. Analysis of the Fate of the N Derived from Fish Feed. Without fish feed application in treatments RF feed 0 and FM feed 0, the N in fish (fish N) was derived from the environment, and the N in rice plants (rice N) was derived from fertilizers and the environment.

With fish feed application in treatments RF feed 1 and FM feed 1, fish N was derived from fish feed and the environment, and rice N was derived from fish feed, fertilizers, and the environment.

To identify the fate of N derived from fish feed in treatments RF feed 1 and FM feed 1, we performed eight calculations. These calculations included the following variables:

fish-*n*, N in fish;
rice-*n*, N in rice;
X_{RF}, N input in fish feed for RF;
X_{FM}, N input in fish feed for FM;
$fish_{RF}$-N1, fish N in RF feed 1;
$rice_{RF}$-N1, rice N in RF feed 1;
$fish_{RF}$-N0, fish N in RF feed 0;
$rice_{RF}$-N0, rice N in RF feed 0;
$fish_{FM}$-N1, fish N in FM feed 1;
$fish_{FM}$-N0, fish N in FM feed 0;
FX_{RF}, fish N that was from fish feed N in treatment RF feed 1;
RX_{RF}, rice N which was from fish feed N in treatment RF feed 1;
FX_{FM}, fish N from fish feed N in treatment of FM feed 1;
(%) FX_{RF}, percentage of total fish feed N incorporated into fish biomass in RF feed 1;
(%) RX_{RF}, percentage of total fish feed N incorporated into rice biomass in RF feed 1;
(%) environment RF, percentage of total fish feed N that remained in the environment in RF feed 1;
(%) FX_{FM}, percentage of total fish feed N incorporated into fish biomass in FM feed 1;
(%) environment FM, percentage of total fish feed N that remained in the environment in FM feed 1.

The calculations were as follows:

$$FX_{RF} = (\text{fish-N1}) – (\text{fish-N0}) \quad \textbf{[S1]}$$

$$RX_{RF} = (\text{rice-N1}) - (\text{rice-N0}) \quad \textbf{[S2]}$$

$$FX_{FM} = (\text{fish}_{FM}\text{-N1}) - (\text{fish}_{FM}\text{-N0}) \quad \textbf{[S3]}$$

$$(\%)\ FM_{RF} = (FX_{RF}/X_{RF}) \times 100 \quad \textbf{[S4]}$$

$$(\%)\ RX_{RF} = (RX_{RF}/X_{RF}) \times 100 \quad \textbf{[S5]}$$

$$(\%)\ \text{environment RF} = ((X_{RF} - FX_{RF} - RX_{RF})/X_{RF}) \times 100 \quad \textbf{[S6]}$$

$$(\%)\ FX_{FM} = (FX_{FM}/X_{FM}) \times 100 \quad \textbf{[S7]}$$

$$(\%)\ \text{environment FM} = ((X_{FM} - FX_{FM})/X_{FM}) \times 100 \quad \textbf{[S8]}$$

1. Cabrera ML, Beare MH (1993) Alkaline persulfate oxidation for determining total nitrogen in microbial biomass extracts. *Soil Sci Soc Am J* 57:1007–1012.

2. Bremner JM, Mulvaney CS (1982) Nitrogen–total. *Methods of Soil Analysis, Part 2. Chemical and Microbiological Properties*, eds, Page AL, Miller RH, Keeney DR (American Society of Agronomy, Soil Science Society of America, Madison, WI), 2nd Ed.

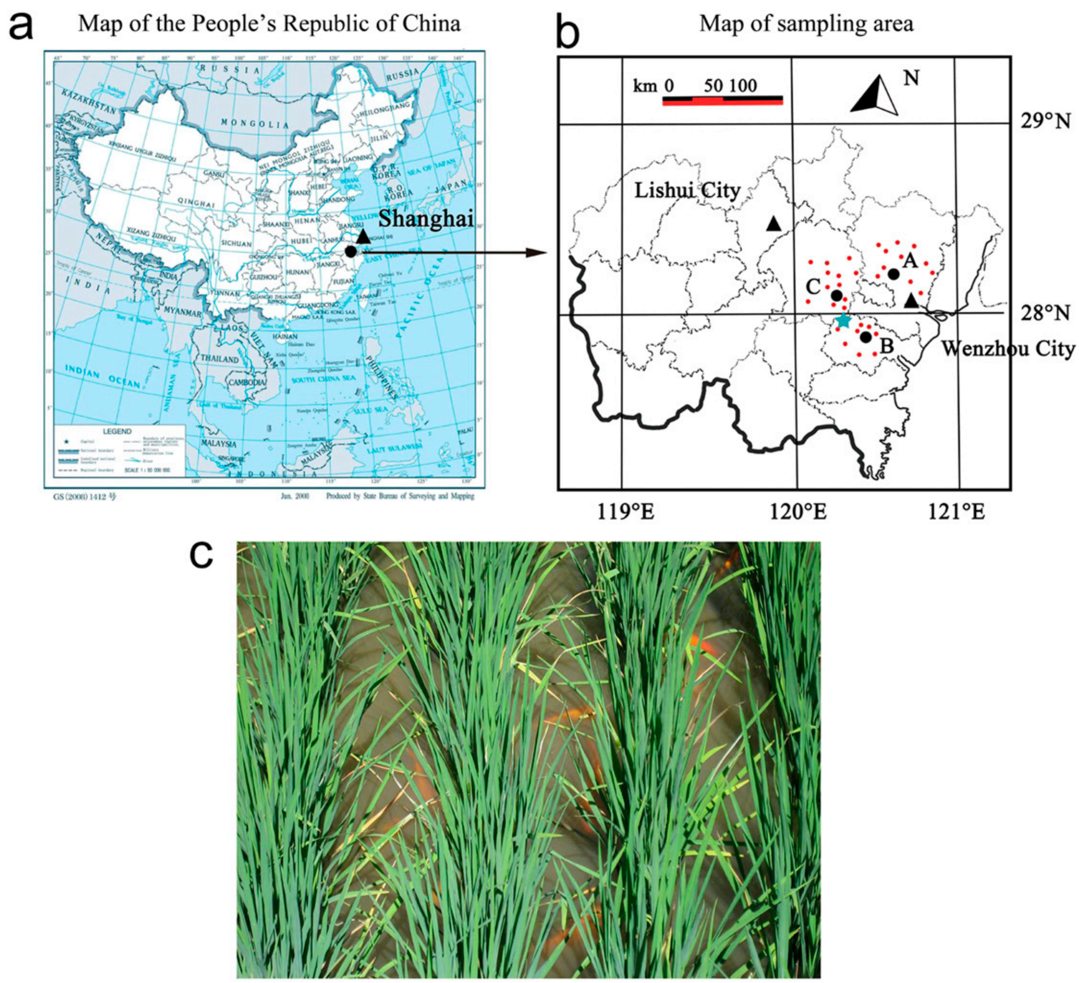

Fig. S1. (*A*–*C*) Map of China (showing the study area) (*A*), map of sampling area (*B*), and picture of rice–fish coculture system (*C*). In the map of China, the black circle indicates the sampling area and the triangle indicates Shanghai city. In the map of the sampling area, red dots indicate the areas sampled in the survey (each included three to five paired subsamples); the green star indicates the GIAHS site, which was the location of the field experiments; the triangles indicate the Lishui City and Wenzhou City; and the three black circles (A, B, and C) indicate the locations of the meteorological stations and monitoring stations for insect pests.

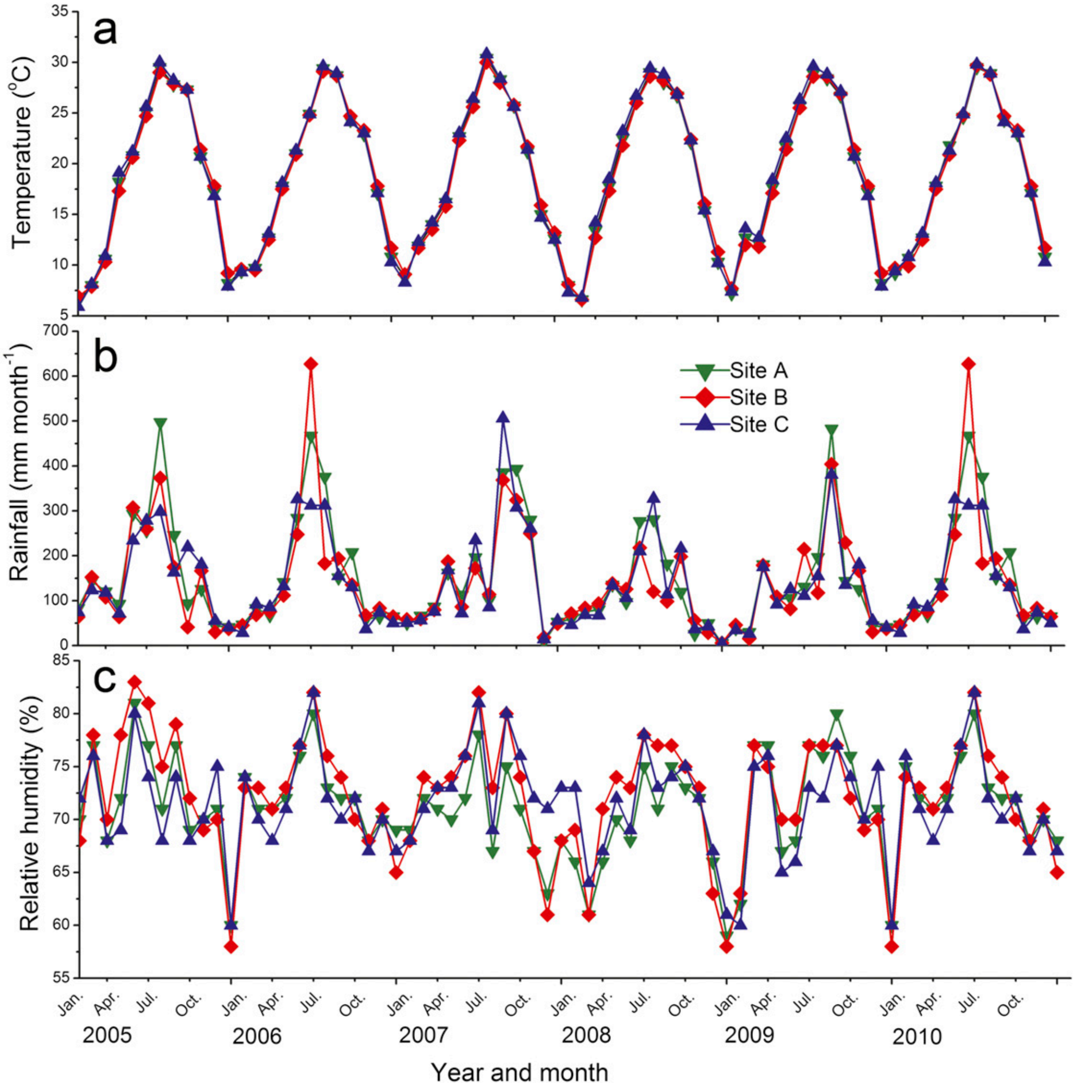

Fig. S2. (*A–C*) Average temperature (*A*), rainfall (*B*), and relative humidity (*C*) in the study area over 6 y of the survey. Data were collected from three meteorological stations (Fig. S1).

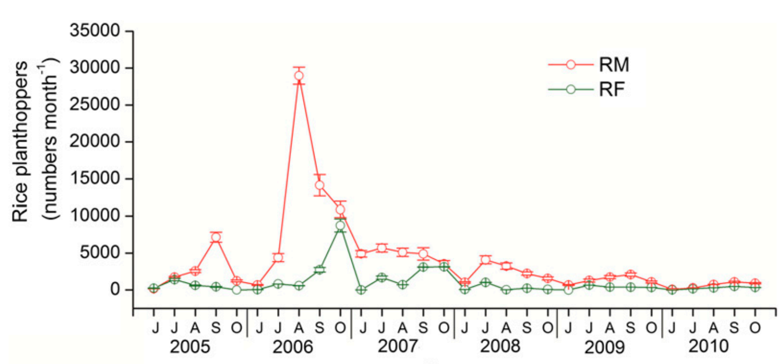

Fig. S3. Dynamics of rice planthoppers in RM and RF over 6 y of the survey. Data were collected from three monitoring stations of rice pests (Fig. S1). Values indicate total catches per month in each year during rice growing season. Values are means for sites A and B for RM and for sites A, B, and C for RF (Fig. S1). RM, rice monoculture; RF, rice–fish coculture. Error bars are SE.

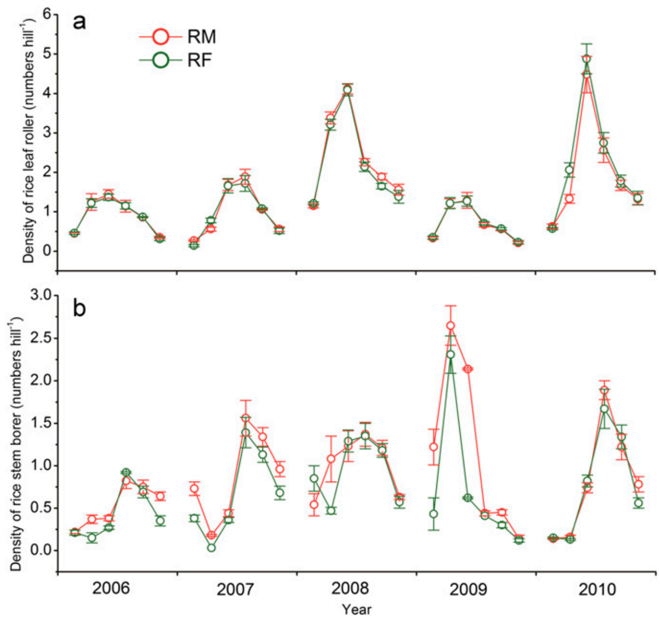

Fig. S4. (*A* and *B*) Abundances of the rice stem borer (*Chilo supperssalis*) (*A*) and the rice leaf roller (*Chaphalocrocis medina*) (*B*) in RM and RF over the 5 y of experiment 1. RM, rice monoculture; RF, rice–fish coculture. Error bars are SE.

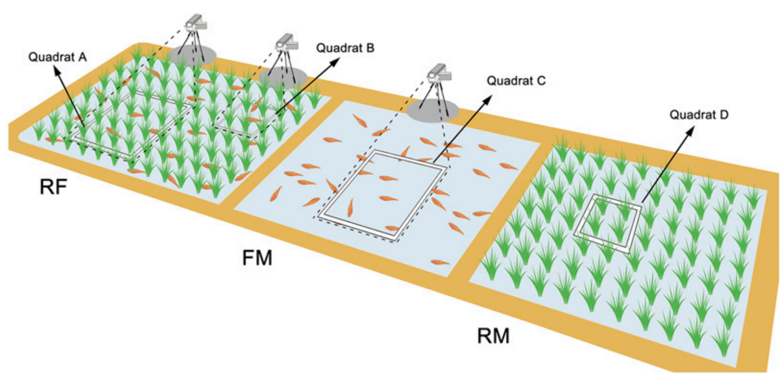

Fig. S5. Diagram indicating how fish activities and rice planthopper movements were recorded in field plots. Quadrat *A* included 16 rice hills, and quadrat *C* was from fish monoculture and included no rice hills; the areas were equivalent for quadrats *C* and *A*. Fish activity in quadrats *A* and *C* was continuously recorded for 3 d with a video camera. Frequency of fish activity was expressed as the number of times a fish appeared in the quadrat in 1 h. In quadrats *B* and *D*, which included 4 rice hills, the surface of the water was covered with rape seed oil, and planthoppers trapped in the oil (they had presumably fallen from the rice plants as a consequence of fish activity, wind, etc.) were removed and counted. Floating bamboo rods, which did not impact fish activity, were used to fence quadrats *A*–*D*. The frequency at which fish hit rice plants (expressed as number of hits per hill per hour) was also determined with recordings of quadrat *B*. RF, rice–fish coculture; FM, fish monoculture; RM, rice monoculture.

Table S1. Fish yield and utilization of pesticides, fertilizers, and fish feeds in rice monoculture (RM) and rice–fish coculture (RF) as determined in the survey

	Variables	RM	RF
Fish yield	Average, kg·ha^{-1}		522.66 ± 80.54
	Coefficient of variation, %		3.66 ± 0.41
Fish feed input	Average, kg·ha^{-1}		1108.13 ± 145.79
	Coefficient of variation, %		1.99 ± 0.13
Pesticides input	Average, a.i. kg·ha^{-1}	14.85 ± 1.50	4.75 ± 0.75
	Coefficient of variation, %	18.80 ± 1.48	11.92 ± 1.49
Total fertilizer input	Average, NPK kg·ha^{-1}	282.27 ± 26.53	215.20 ± 19.30
	Coefficient of variation, %	3.14 ± 0.23	3.72 ± 0.28
Nitrogen fertilizer input	Average, N kg·ha^{-1}	183.54 ± 15.84	149.49 ± 14.51
	Coefficient of variation, %	1.90 ± 0.33	1.67 ± 0.10

Values are means for 2005–2010. Values are means ± SE. a.i., active ingredient; K, potassium; N, nitrogen; P, phosphorus.