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# Discrimination of Newly Immigrated White-Backed Rice Planthoppers, *Sogatella furcifera* (Homoptera: Delphacidae) by Lipid Analysis[1]

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The white-backed planthoppers lose their dry body weight and lipid contents on a long distance flight. We analyzed these variables of insects caught over the East China Sea (31°N, 126°E) and from various parts of the Japanese islands in June and July 1986 and 1987, and compared their discriminant functions. Mean numbers of newly immigrated planthoppers, so defined when they showed the same levels of variables as those of the East China Sea population, were 4.2 to 4.5 out of 12 analyzed in samples from Nagasaki and elsewhere in Japan. They were always found in association with the synoptical frontal systems developed over the Japanese Islands. This finding was in conformity with the previously proposed migration theory. Significantly different dry body weights and triglyceride and fatty acid contents among the East China Sea populations caught in 1986 and 1987 suggested that their emigration origins differed.

## INTRODUCTION

The white-backed planthopper, *Sogatella furcifera* HORVÁTH, and the brown planthopper, *Nilaparvata lugens* STÅL, are major pests of rice plant in East Asia. On their occurrence in Japan there have been two theories proposed: immigration from overseas (KISIMOTO, 1975) and overwintering (TAKEZAWA, 1961). Since many planthoppers were found gathering at a night light of a weather ship anchored 500 km south of mainland Japan on the Pacific Ocean (ASAHINA and TSURUOKA, 1968), the immigration theory has gained momentum. Systematic collections of insects immigrating from overseas have been made over the East China Sea, and thousands of planthoppers are caught every year (KISIMOTO, 1981; OGAWA, 1987). In addition, outbreaks of planthoppers in Okinawa and Kyushu were significantly correlated with the numbers caught on the East China Sea (KISIMOTO, 1981).

Flight capability of planthoppers was proven by researchers using a flight mill.

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For example, flight duration of the brown planthopper extended from a few sec to more than 20 hr (OHKUBO, 1973; PADGAM, 1983a). These insects also had a large amount of lipds as flight fuel (PADGAM, 1983b).

Thus the possibility of their immigration to Japan is very high, yet there is no direct proof of it. In this work we tried to discriminate newly immigrated white-backed planthoppers from those previously settled and discussed their immigration in relation to synoptic conditions.

## MATERIALS AND METHODS

*Insects.* Macropterous females of the white-backed planthopper, *Sogatella furcifera* HORVÁTH, caught in the following fields on the dates shown were used.

a. East China Sea populations: Insects were caught with net traps on the weather observation ship KEIFUUMARU stationed near a point 31 °N, 126 °E (midway between Shanghai and Kyushu) on June 29, 1986 and July 7, 10 and 11, 1987. Insects caught on the respective dates were respectively numbered 1 to 12, 13 to 24, 25 to 36 and 37 to 48 for individual analysis.

b. Nagasaki populations: Insects were caught with light traps at Nagasaki Experimental Station in Isahaya City, Kyushu. Only those caught on days when more than 24 females were trapped were analyzed. There were 17 such days from June 17 to July 19 in 1986, and in 1987, 12 days including June 2 and 8, and every even numbered day from July 2 to 20.

c. Populations from various parts of Japan: Insects were caught by light traps in 36 prefectures from June 6 to Sep. 10, 1985 and around June 18, 30 and July 15, 1986 in 25 prefectures. The samples in which there were more than 24 females were analyzed.

All the insects were preserved in a desiccator until use.

*Measurement of dry body weight and forewing length.* Dry body weight (DW) of an insect was determined with a balance (Shimadzu L-200SM, threshold of 0.01 mg). A forewing was removed from the thorax and the maximum length was measured with a micrometer under a microscope. Twelve insects were analyzed for each sample except the 1985 samples, in which 6 of each were analyzed.

*Measurement of lipid contents.* After measuring dry weight and wing length, the insect was dipped into 0.2 ml of a solvent consisting of an equal volume of chloroform and acetone in a 2-ml glass vial and kept in a refrigerator overnight. The insect was macerated with the tip of a glass capillary and the solution was dropped onto a 25×25 cm silicagel plate (Wako Chemical Co.). The plate was developed with a solvent of hexane: ether: acetic acid. 80: 20: 2. Developed plates were sprayed with 50–60% sulfuric acid using an auto spray (Shimadzu SPU-1) and charred in an oven at 130–140°C for about 15 min. The spots were scanned with a chromato scanner (Shimadzu CS-920). Similarly, known amounts of authentic triolein and oleic acid were chromatographed for calibration curves. In a preliminary analysis of the insects, hydrocarbon, cholesterol, cholesterol ester, diglycerides and monoglyceride were detected in addition to triglyceride (TG) and fatty acid (FA). However, only the latter two were measured because of their significant amount.

*Discrimination of new immigrants.* The 3 samples of East China Sea populations were divided into 3 groups based on body density index (dry weight in mg divided by triple wing length in mm and multiplied by 100). TG and FA contents (μg/insect) by hierarchial clustering (single linkage method). Then, discriminant functions were calculated between the East China Sea groups and each Japanese population to determine if the latter individuals belonged to the former groups. If so, they were considered as new immigrants. Here, new immigrants are defined as those having landed within the last few days because four days after landing is required for full resynthesis of lipid (PADGAM, 1983a) and recovery of body weight (INOUE et al., 1988). A personal computer (Nihon Electric Co. 9801–VM29) and the program MULVA 2 were used for calculation.

## RESULTS

*Analysis of the East China Sea populations*

The 1986 population had smaller mean values of dry weight body density index (BD) and lipid contents than the 1987 populations (Table 1). Only one individual from the 1986 sample contained any TG, in spite of their similar forewing lengths. There were some differences among the 1987 populations, i.e. DW, BD and TG of the July 11 populations were somewhat larger than those of the others. By hierarchial clustering all the 48 insects could be divided into 3 groups. Twenty-two insects from No. 1 to

Table 1. Mean dry body weight and lipid contents of 4 female *S. furcifera* populations caught on the East China Sea in 1986 and 1987

| Date | DW (mg±SD) | WL (mm±SD) | BD (mg/mm³±SD) | TG (μg±SD) | FA (μg±SD) |
|---|---|---|---|---|---|
| 1986.6.29 | 0.38±0.06 | 3.67±0.13 | 0.78±0.13 | 2.4± 8.1 | 18.3±13.8 |
| 1987.7. 7 | 0.44±0.08 | 3.67±0.13 | 0.89±0.21 | 18.2±17.0 | 59.4±16.7 |
| 7.10 | 0.46±0.11 | 3.62±0.20 | 0.97±0.21 | 13.4± 9.7 | 40.6±21.9 |
| 7.11 | 0.55±0.12 | 3.75±0.14 | 1.03±0.20 | 43.0±35.0 | 40.9±26.7 |

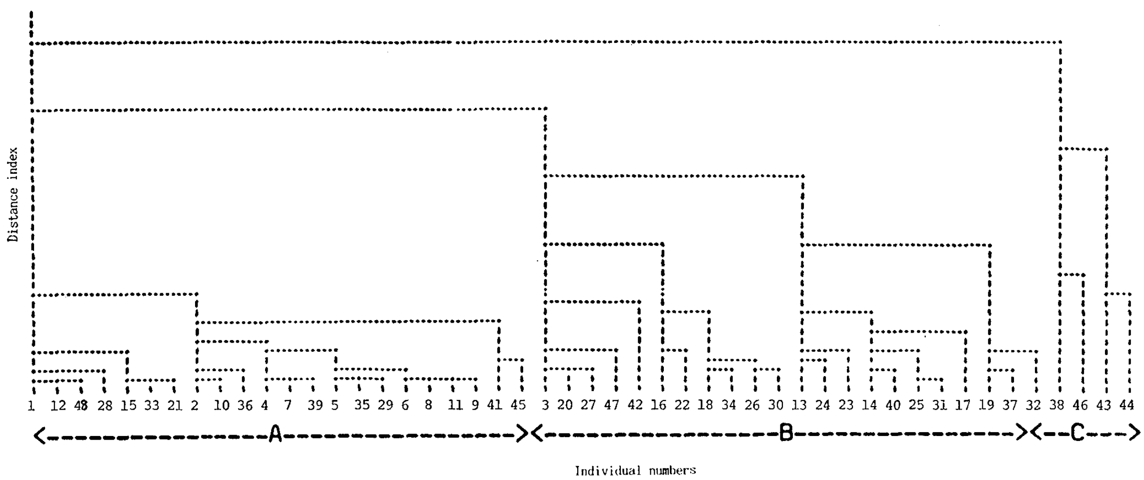


Fig. 1. Hierarchial clustering of 48 female *S. furcifera* caught over the East China Sea. Insects numbered 1 to 12, 13 to 24, 25 to 36 and 37 to 48 were caught on 1986-6-29, 1987-7-7, 7-10 and 7-11, respectively.

Table 2. Mean dry body weight and lipid contents of groups A, B and C of female *S. furcifera* caught on the East China Sea in 1986 and 1987

| Group | *n* | DW (mg±SD) | WL (mm±SD) | BD (mg/mm$^3$ ×100±SD) | TG (μg±SD) | FA (μg±SD) |
|---|---|---|---|---|---|---|
| A | 22 | 0.38±0.05 | 3.65±0.14 | 0.78±0.12 | 1.4± 4.4 | 17.1± 7.4 |
| B | 22 | 0.51±0.10 | 3.67±0.16 | 1.04±0.20 | 25.1±17.4 | 61.1±12.2 |
| C | 4 | 0.60±0.08 | 3.86±0.53 | 1.05±0.13 | 85.0±11.5 | 47.6±29.1 |

Table 3. Numbers of female *S. furcifera* of the East China Sea populations divided into 3 groups

| Date | Populations | | |
|---|---|---|---|
| | A | B | C |
| 1986.6.29 | 11 | 1 | 0 |
| 1987.7. 7 | 2 | 10 | 0 |
| 1987.7.10 | 5 | 7 | 0 |
| 1987.7.11 | 4 | 4 | 4 |

Table 4. Mean dry body weight and lipid contents of female *S. furcifera* trapped at Nagasaki Experimental Station during the immigrating season in 1986 ($n=12$ each)

| Date | DW (mg±SD) | WL (mm±SD) | BD (mg/mm$^3$ ×100±SD) | TG (μg±SD) | FA (μg±SD) |
|---|---|---|---|---|---|
| Jun. 17 | 0.46±0.07 | 3.70±0.15 | 0.90±0.13 | 12.8±13.2 | 49.1±17.2 |
| 18 | 0.54±0.11 | 3.77±0.10 | 1.02±0.20 | 19.6±16.9 | 50.6±21.7 |
| 19 | 0.58±0.05 | 3.75±0.12 | 1.11±0.18 | 16.0±13.7 | 93.3±16.0 |
| 21 | 0.42±0.06 | 3.72±0.20 | 0.82±0.10 | 5.8± 9.7 | 32.6±14.8 |
| 22 | 0.50±0.08 | 3.77±0.17 | 0.89±0.10 | 4.5± 6.4 | 42.0±23.1 |
| 23 | 0.45±0.08 | 3.77±0.17 | 0.84±0.13 | 4.0± 3.9 | 41.8±22.4 |
| 25 | 0.55±0.08 | 3.80±0.14 | 1.00±0.15 | 20.9±35.1 | 54.0±28.4 |
| 29 | 0.48±0.03 | 3.70±0.08 | 0.96±0.10 | 18.7±13.7 | 55.9±12.7 |
| 30 | 0.51±0.08 | 3.66±0.11 | 1.03±0.17 | 24.9± 9.2 | 51.4±15.2 |
| 30 | 0.46±0.07 | 3.72±0.12 | 0.90±0.16 | 12.8±12.3 | 39.7±12.3 |
| Jul. 1 | 0.55±0.12 | 3.78±0.16 | 1.01±0.17 | 7.1± 7.4 | 61.7±20.4 |
| 5 | 0.47±0.05 | 3.69±0.14 | 0.93±0.11 | 19.1±15.6 | 47.7± 7.7 |
| 6 | 0.54±0.13 | 3.73±0.14 | 1.04±0.23 | 7.2±15.0 | 74.6±17.8 |
| 7 | 0.45±0.06 | 3.76±0.09 | 0.84±0.12 | 9.6±13.5 | 41.7±18.5 |
| 13 | 0.52±0.08 | 3.79±0.13 | 0.97±0.19 | 44.7±41.9 | 80.4±21.8 |
| 15 | 0.47±0.09 | 3.75±0.10 | 0.88±0.12 | 61.5±13.1 | 71.1±14.8 |
| 17 | 0.47±0.06 | 3.65±0.16 | 0.96±0.09 | 19.7±12.0 | 59.6±17.5 |
| 18 | 0.47±0.06 | 3.73±0.09 | 0.90±0.11 | 4.3± 6.0 | 39.8±12.7 |
| 19 | 0.65±0.12 | 3.78±0.14 | 1.20±0.23 | 36.6±31.0 | 76.4±18.8 |

No. 45 were defined as group A, 22 from No. 3 to No. 32 and 4 from No. 38 to No. 44 as group B and C, respectively (Fig. 1). Group A had the lowest values of all, particularly in TG content (Table 2). On the contrary, group C had the highest values except in FA content. The TG content of group C was 85.0 μg, which was 60 and 3.4

Table 5. Mean dry body weight and lipid contents of female *S. furcifera* trapped at Nagasaki Experimental Station during the immigrating season in 1987 ($n$=12 each)

| Date | DW (mg±SD) | WL (mm±SD) | BD (mg/mm³ ×100±SD) | TG (μg±SD) | FA (μg±SD) |
|---|---|---|---|---|---|
| Jun. 2 | 0.47±0.08 | 3.73±0.15 | 0.91±0.17 | 25.1±28.6 | 47.5±15.0 |
| 8 | 0.41±0.05 | 3.78±0.12 | 0.76±0.08 | 0.0± 0.0 | 34.5±13.0 |
| Jul. 2 | 0.45±0.09 | 3.67±0.12 | 0.90±0.15 | 30.6±26.0 | 56.1±14.6 |
| 4 | 0.46±0.11 | 3.70±0.11 | 0.90±0.20 | 8.8±14.3 | 29.6±18.0 |
| 6 | 0.58±0.15 | 3.72±0.17 | 1.10±0.26 | 26.6±41.8 | 53.7±23.6 |
| 8 | 0.64±0.13 | 3.72±0.13 | 1.25±0.19 | 20.5±20.5 | 87.7±16.4 |
| 10 | 0.51±0.15 | 3.75±0.09 | 0.97±0.29 | 10.3±19.5 | 47.7±20.7 |
| 12 | 0.76±0.18 | 3.79±0.15 | 1.38±0.29 | 2.9± 4.1 | 118.1±40.5 |
| 14 | 0.66±0.15 | 3.79±0.09 | 1.21±0.25 | 9.7±15.7 | 90.7±36.6 |
| 16 | 0.42±0.10 | 3.73±0.09 | 0.80±0.16 | 11.8±17.7 | 62.0±23.2 |
| 18 | 0.45±0.12 | 3.69±0.15 | 0.87±0.16 | 6.5±14.2 | 64.4±28.9 |
| 20 | 0.45±0.13 | 3.72±0.10 | 0.88±0.23 | 20.2±28.6 | 80.3±41.4 |

times as large as those of group A and B, respectively. The majority of the 1986 population, 11 out of 12 were in group A, whereas the 1987-7-11 population was evenly distributed into 3 groups (Table 3). The remaining two were grouped in A and B.

*Analysis of the Nagasaki populations*

Among the 5 parameters of the Nagasaki populations, the mean TG contents were the most variable and showed the largest standard deviations (Tables 4 and 5): the minimum and maximum contents of the 1986 populations were 4.0 μg and 61.5 μg, respectively. The mean FA content was also variable, showing a maximum of 93.3 μg and a minimum of 32.6 μg. Similarly, the mean lipid contents of the 1987 populations were variable. These facts imply that these populations are highly heterogenous ones composed of new immigrants and previously settled insects.

Each insect was, therefore, discriminated to see if it belonged to any of the East China Sea populations (immigrants). In the 1986 populations, 1.6, 2.7 and 0.2 insects out of 12 analyzed were determined to belong to group A, B and C, respectively. The total numbers of immigrants (all groups combined) varied from 0 to 9 depending on the dates trapped and the average was 4.5 (Fig. 2). However, no significant correlation was observed between the numbers trapped and those newly immigrated ($r$=0.273, $n$=19). In the 1987 populations, the mean numbers of groups A, B and C and the total were 2.3, 2.3, 0.3 and 4.5, respectively, which were similar to those of the previous year (Fig. 3). Again no significant correlation was observed between the numbers trapped and newly immigrated ($r$=0.120, $n$=12).

*Analysis of other Japanese populations*

Number of new immigrants out of 6 females in each sample caught over the Japanese islands in 1985 were from 0 to 4, of which the average was 1.2. Out of 227 insects analyzed, 24 belonged to group A, 17 to group B and only 2 to group C. In 16 prefectures there were no new immigrants. The only new immigrants were seen on July 13, 14 and 15 on which dates the analytical data show the locations of front systems developing (Fig. 4). Clearly, they were distributed in the southern part of the 13N

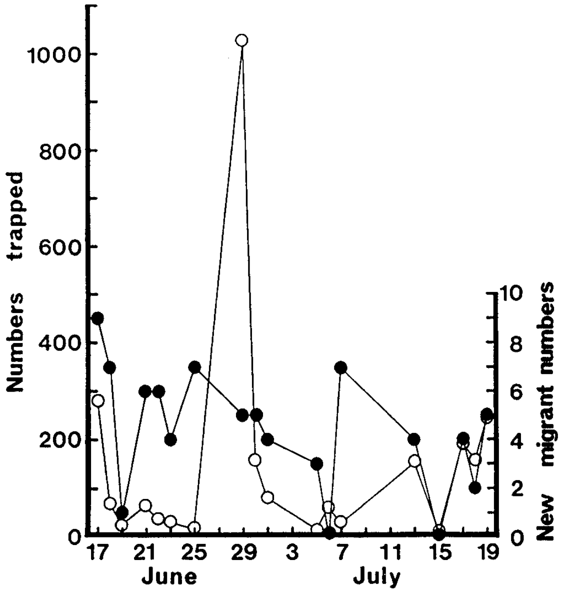


Fig. 2. Numbers of female *S. furcifera* caught by light traps (open circles) and new immigrants (solid circles) in Nagasaki Experimental Station in 1986. New immigrants were defined as insects having the same characteristics as those caught on the East China Sea.

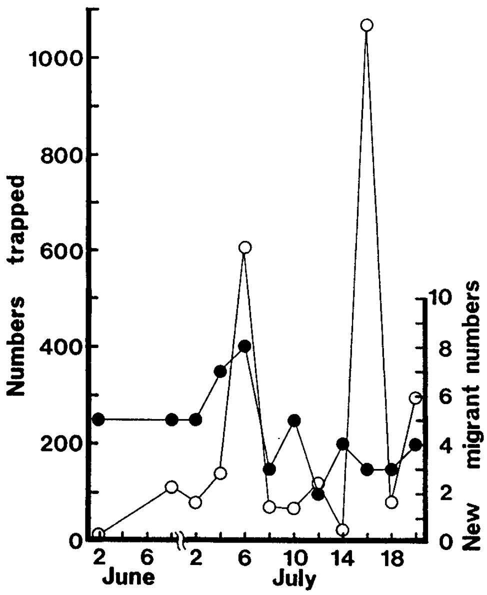


Fig. 3. Numbers of female *S. furcifera* caught by light traps (open circles) and new immigrants (solid circles) in Nagasaki Experimental Station in 1987.

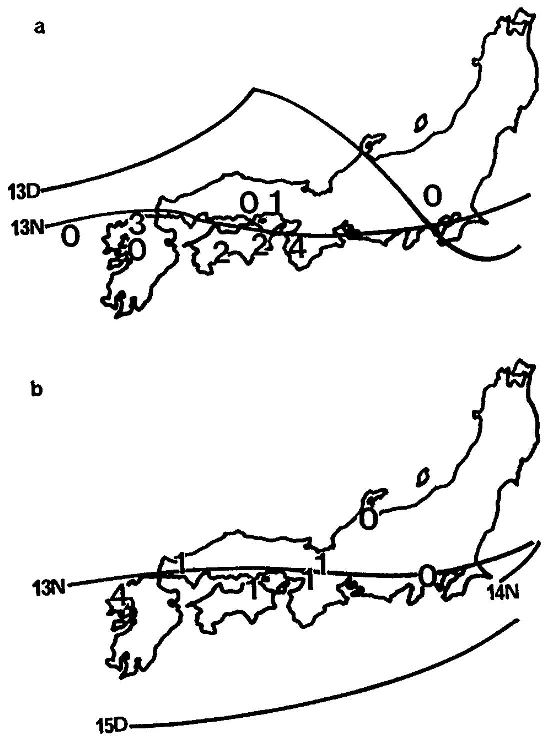


Fig. 4. Location of the frontal systems over the Japanese islands and new immigrant numbers out of 6 female *S. furcifera* analyzed in each prefecture in 1985. "a" for July 13 and "b" for July 14/15. "D" and "N" for the fronts at 9:00 and 21:00 respectively.

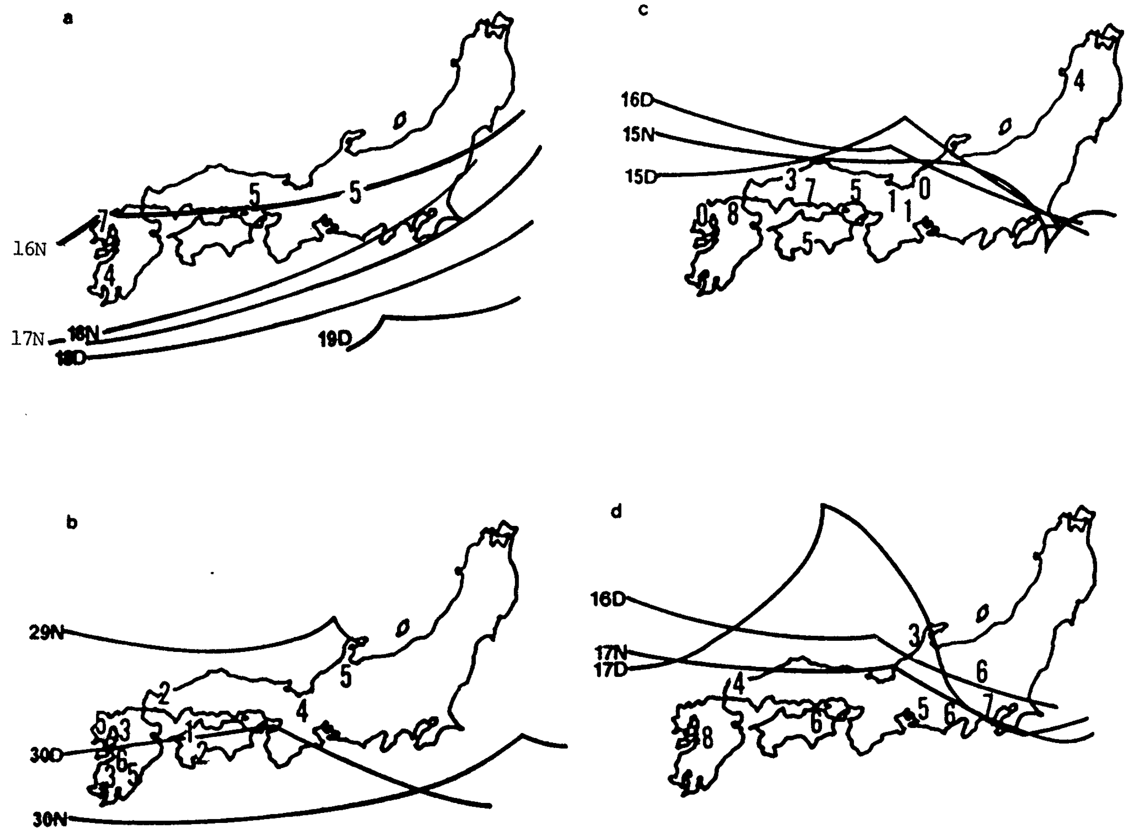


Fig. 5. Location of frontal systems over the Japanese islands and new immigrants from among 12 female *S. furcifera* analyzed in each prefecture in 1986. "a" for June 18/19, "b" for June 29/30, "c" for July 15, and "d" for July 16/17. "D" and "N" for fronts at 9:00 and 21:00, respectively.

(at night of the 13th) frontal system. Therefore, it is deduced that a large wave of new immigrants landed on the night of July 13.

In 1986, the minimum and maximum numbers of new immigrants among 12 females analyzed each date were 0 and 8, respectively, and the average was 4.2 (Fig. 5). Only 2 prefectures had no new immigrants, but 9 prefectures had more than 6. Out of all 151 new immigrants, 52, 80 and 19 belonged to group A, B and C, respectively. Immigration was again well associated with the frontal systems (Fig. 5). The immigrants of the June 18 and 19 populations could be related to the 16N frontal system which had passed 2 days earlier.

## DISCUSSION

*Definition of new immigrants and their proportion in light trap samples*

Discrimination of different insect populations by analysis of metal contents was done by BOWDEN et al. (1979) with *Noctua pronuba* and TURNOCK et al. (1979) with *Entomoscelis americana*. For discrimination of new immigrants in the light trap populations of the rice planthoppers, INOUE et al. (1988) used Ca and Mg contents in addition to body density index as markers. Lipid contents (and body density index) were used in the present work. In the preliminary analysis of lipids, the 1984 and 1985 insects caught on the East China Sea retained no lipids, apparently all lipid fuel had been exhausted on their arrival there. Meanwhile, PADGAM (1983b) showed that it takes 4 days for complete resynthesis of lipids after insect flight under laboratory conditions. In this connection INOUE et al. (1988) also showed a 4 day recovery period was necessary for dry body weight of new immigrants to Kyushu, when bred on rice plants in cages. Therefore, new immigrants can properly be defined as being within 4 days after arrival.

Although planthoppers caught by light traps are thought to have immigrated (PERFECT et al., 1985), not all insects trapped in the present work were new immigrants, and the numbers trapped could not be significantly correlated with those of new immigrants (Figs. 2 and 3). The reason may be that some previously established, lipid-synthesizing insects were trapped while taking a short trivial flight. In fact, a significant correlation seems to exist between these numbers in the initial phase of their immigration, although it becomes insignificant with mounting numbers of previous immigrants (Figs. 2 and 3). Similar insignificant correlation was observed when new immigrants were determined by the measurement of decrease in Ca and Mg content, which recovered within a day of their arrival (INOUE et al., 1988).

*Divergence of immigrant sources*

Insects in group A caught on the East China Sea had very small dry weight and little lipid content, while those in group C had more (Table 2). This implies a different distance and duration of their flight. In other words, group A flew a longer time and a greater distance than group C. Since the insects caught over the East China Sea were thought to originate from China, Taiwan and Okinawa (ROSENBERG and MAGOR, 1983), the origins of group A and C might be South China and Okinawa, respectively. The 1986–6–29 population in which most belonged to group A is thought to have immigrated from the southern part of China. On the contrary, the 1987–7 populations would be from various sources from Okinawa to China. The 1987–7–11 population was caught in an unusual southerly wind when a typhoon was approaching from east of Taiwan. Insects from different sources on the same wind path can be caught simultaneously (ROSENBERG and MAGOR, 1983). However, immigrant origins should be determined by analyses of other biochemical or genetical characteristics of emigrating strains from possible locales in future.

*New immigrants in relation to frontal systems*

The present finding was in accordance with the previous fact in which numbers in light trap catches and location of front systems are closely related (KISIMOTO, 1976). The new immigrants in the 1985–7–14/15 population might have immigrated on July 13, 1 or 2 days earlier than they were trapped (Fig. 4). Those in the 1986–6–18 population are also presumed to have landed on June 16 when the frontal system stayed over Honshu Island (Fig. 5a). These discrepancies between development of a frontal system and detection of new immigrants may be justified since insects take a few days to resynthesize lipids following arrival (PADGAM, 1983a).

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