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Effects of the Inabenfide [4'-chloro-2'-(α-hydroxybenzyl)-isonicotinanilide] on the Growth of Rice Grown in No Tillage Cultivation with Single Basal Fertilization to the Nursery Box

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ABSTRACT

Field experiments were conducted to determine the effects of the inhibitor of gibberellin (GA) biosynthesis on the growth and yield of rice (*Oryza sativa* L.) grown by no tillage cultivation with single basal fertilization of Controlled-Availability Fertilizer (CAF) to the nursery box. Inabenfide (4'-chloro-2'-(α-hydroxybenzyl)-isonicotinanilide: IBF) was used as an inhibitor of GA biosynthesis. There was no difference in the plant age between the control and IBF treatment. However, the length of IBF-treated plants is significantly longer than the control plants during the growth period. The number of tillers was similar between both treatments until the maximum tiller number stage, after that, it significantly increased in IBF-treated plants. The IBF treatments increased the number of panicles by 24.5% compared with the control but decreased the number of grains per panicle. The IBF treatment did not affect the total number of grains, the reduction in grain number per panicle was compensated by the increased number of panicles. The percentage of ripened grains and the one-thousand-grain weight in IBF-treated plants were similar to those of the control. When all yield components were taken into consideration, no difference in grain yield was found between the IBF treatment and control groups. The present report indicates that IBF works well for promoting tillering in no-tillage rice cultivation with a single basal fertilization of CAF to the nursery box.

Key words: Controlled availability fertilizer, inabenfide, inhibitor of gibberellin biosynthesis, no-tilled cultivation, rice, tiller

INTRODUCTION

Rice is one of the staple food crops in Asia. Recently, many attentions have been paid on establishment for the rice cultivation which require less labor and time as well as eco-friendly. Single basal applications of Controlled-Availability Fertilizer (CAF) to nursery boxes in no-tillage rice cultures is effective for saving labor, reducing production costs and achieving environment-friendly farming (Kaneta, 1995; Saigusa, 1999; Wakimoto, 2004; Watanabe *et al*., 2006). However, a problem with this culture is delayed early growth, especially a reduction of tillering which results in reduced panicle numbers per square meter and grain yields (Takahashi, 1993; Ando *et al*., 1998; Ito, 2002).

There are many growth retardants currently available for restricting growth and the most commonly used and best understood group inhibits gibberellin (GA) biosynthesis (Arteca, 1996;

Gianfagna, 1995). Among the many GA biosynthesis inhibitors is inabenfide (IBF), an isonicotinic acid anilide derivative which inhibits GA biosynthesis by blocking the oxidative pathway (Arteca, 1996; Gianfagna, 1995; Miki *et al*., 1990). In rice cultivation, inhibitors of GA biosynthesis are known to have an inhibiting effect on lodging. However, the compound has been known have the side effect of promoting tillering in some cases (Shirakawa, 1990; Fukazawa and Shirakawa, 2001). Therefore, we investigated the effect of IBF on growth and yield of rice, with special attention to tillering and panicle number, when grown in no-tillage cultures with a single basal application of controlled availability fertilizer to the nursery box.

MATERIALS AND METHODS

Varieties and cultivation method: The field trial was conducted in 2007 at the Field Science Center of Tohoku University, Miyagi Prefecture, Japan (38°44' N., 140° 15' E., alt. 220 m). The cultivar used in the study was Hitomebore (*Oryza sativa* L.). The soil was a typical non-allophane volcanic ash soil (Andisol, Alic Pachic Mellanudand). Two kinds of CAF, polyolefin coated urea 30 (POCU30) and polyolefin coated urea s100 (POCUs100) were used; the POCU30 releases 80% of its nitrogen content during 30 days in water at 25°C (Shoji and Gandeza, 1992). The POCUs100 has 30 days of suppressed release and 70 days of sigmoid release. The sigmoid type of POCUs100 was, as a source of CAF, used as a single basal application of 5 g m^{-2} total nitrogen in a nursery box at the time of sowing (Fig. 1). This fertilization method enables to contact application of seeds and fertilizer without roots injuries and thus it eliminate the need to top-dress. The linear type of POCU 30 was used at the rate of 2 g m^{-2} broadcast in the nursery box one day before transplanting to improve the initial growth of rice. The plant density was 22.2 hills m^{-2} (30×15 cm). The seedlings (4.0-4.5 plant age, 4 plants per hill) were transplanted by a non-tillage transplanter (Mitsubishi Co., Japan) (Fig. 2). No-tillage fields were sprayed with 4.1 L a.i. ha^{-1} of isopropyl ammonium glyphosate to control weeds at 20 days before transplanting. The experimental field was submerged in water 10 days before transplanting. Rice straw was scattered on the surface of soil. The transplanting date was May 11th. The granular formations containing 4% of IBF were applied to the nursery box (60×30×15 cm) at the rate of 75 g per box (0.6g a.i. m^{-2}) by hand one day before transplanting. The no-tillage paddy field was divided into two regimes, control (untreated IBF) and IBF treatments. Both fields were 7.2×30 m (24 rows×30 m) in size. Three plots were

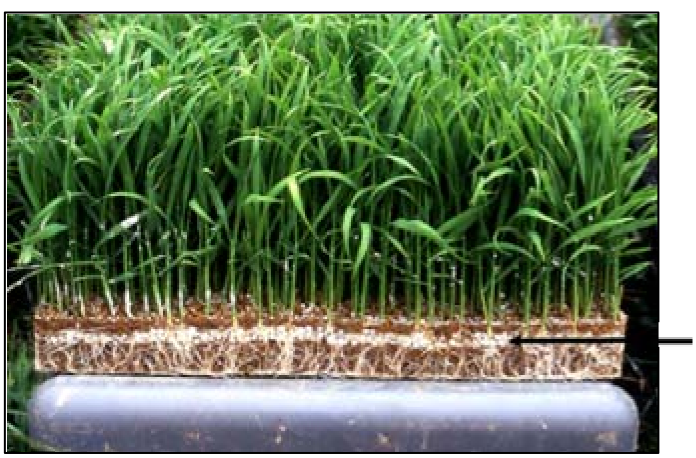

Fig. 1: Single basal applications of rice seed and Controlled-Availability Fertilizer (CAF) to nursery boxes. The black arrow indicates the particle of CAF

Fig. 2: Transplanting by a non-tillage transplanter

randomly selected from both the IBF treatment and the control. Each plot consisted of ten plants (2 rows×5 hills in quadrant).

Morphological traits of rice plants: The plant age, plant length, leaf color (SPAD value) and tiller number of each plot (10 hills) were investigated. In rice, the leaf number on the main stem is used as an indicator of plant age and as a time scale of the growth. The plant age is expressed by the number of emerged leaves and the ratio of the visible length of an emerged leaf to its final length (Hoshikawa, 1989). For example, a plant age of 6.5 means that 6 leaves are fully emerged and the seventh leaf has emerged to half of its final length. The decimal fraction of the plant age changes continuously because there is only one emerging leaf in the rice plant. Leaf color was measured using a SPAD-502 chlorophyll instrument (Konica Minorta Sensing Inc, Japan).

Dry matter production: At the maximum tiller stage and harvest time, three batches of averaged sample (one batch consists of 2 hills) from three plots of each treatment were taken and separated into leaf blade, leaf sheath, stem, dead leaves and panicle. The dry weight of each sample was determined after oven drying at 70°C for 72 h to a constant weight.

Grain yield and yield components: Grain yield and yield components were investigated by standard methods (Yoshida, 1981) with slight modifications. Prior to harvest, five plants which showed the average number of panicles were obtained from each plot of controls and IBF treatments to measure yield components. The number of panicles in each sample was counted. The grains were divided into filled grains and unfilled grains using salt water with a gravity of 1.06 and the percentages of filled grain were determined. The brown rice one-thousand-grain weight was measured and grain weight was calculated at 15% moisture content. For analysis of grain yield, three batches of 24 hills were collected (One batch consists of 6 rows×4 hills per quadrant).

Statistical analysis: Mean and Standard Error (SE) of three plots from IBF treatments and controls were calculated, respectively. The mean values were compared by Student's t-test (n = 3) at the 5 and 1% levels.

RESULTS AND DISCUSSION

Air temperature condition: The air temperature during rice growth period in 2007 is shown in Fig. 3. The temperature in 2007 coursed similarly with that of average year until the end of June, the duration includes early growth and the maximum tiller number stages. Thereafter, the temperature in 2007 moved on higher than that of average year until the end of September, the period consists of the panicle initiation, heading and ripening stages.

Morphological traits of rice plants: The effects of IBF on growth characteristics of rice plants are shown in Fig. 4. There was no difference in the plant age between the IBF-treated plants and

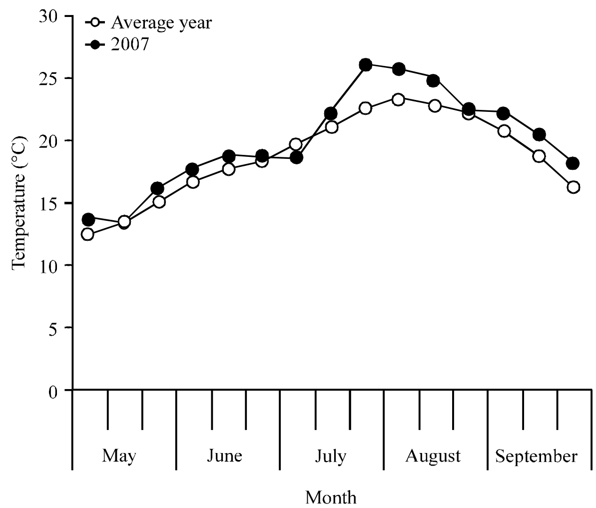

Fig. 3: Air temperature conditions in experiment year

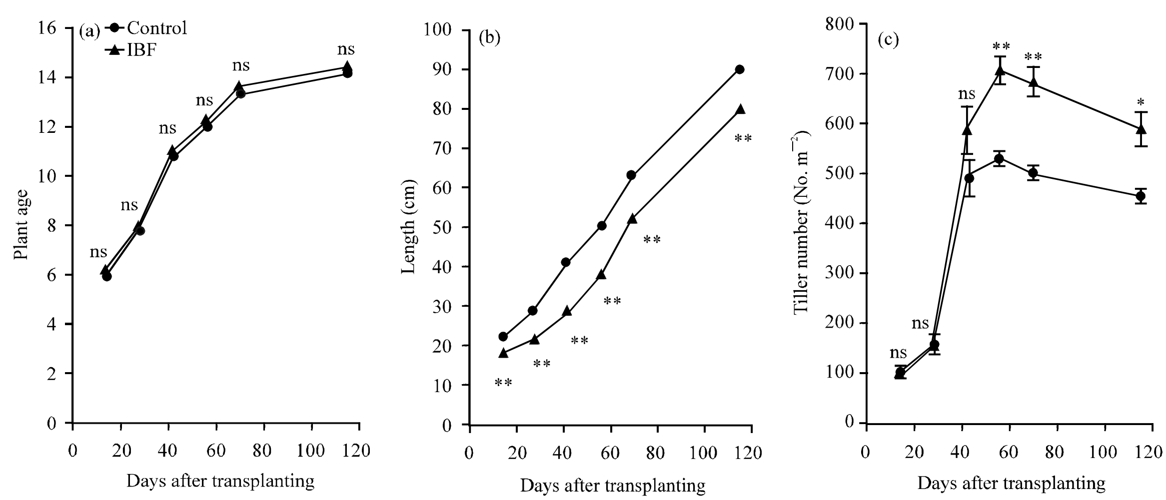

Fig. 4(a-c): Changes in (a) Plant age, (b) Plant length and (c) Tiller number with or without IBF (control). Bars show Means±SE (n = 3). *,**: Significant difference compared the control at the 5 and 1% levels, respectively. ns: No statistical difference between the control and IBF treatments

Fig. 5: Effects of IBF treatment on shoot growth of rice at the maximum tiller number stage, Left to right: Control and IBF treatment

the controls. However, the length of IBF-treated plants is markedly shorter than those of the control plants during the growth period. There was no difference between the IBF and control plants in tiller numbers for the first 42 days after transplantation. By day 56 both groups had reached their maximum tiller number stages but the number of tillers in the IBF-treated plants had increased significantly over those of the controls. Even though the length of IBF-treated plants was shorter than that of control plants, the maximum tiller number was increased by IBF treatments (Fig. 5).

Dry matter production: The dry weight of whole shoot and leaf sheath plus culm in IBF plots at maximum tiller number stage were significantly less than those of control plots, whereas, differences in the dry weight of shoots were not observed at harvest (Fig. 6). This means that IBF affects the dry matter production from early stage to maximum tiller number stage but does not affect dry matter production at the yielding stage.

Grain yield and yield components: Table 1 shows the comparison of grain yield and yield components of rice between the IBF-treated and control plants, IBF treatments significantly increased the number of panicles by 24.5% compared with the control but reduced the number of grains per panicle. The IBF did not affect the total number of grains (control, 27418±1560 grains m^{-2}, IBF; 25775±1545 grains m^{-2}), thus the reduction for grain number per panicle was compensated by the number of panicles. The percentage of ripened grains and 1000 grain weight was not affected by IBF treatments. When all yield components were taken into consideration, no difference in grain yield was found between IBF treatments and control.

In monocotyledonous plant, exogenously applied kinetin, a cytokinin, is known to increase tiller growth and the percentage of tiller-bud release in wheat (Langer *et al*., 1973) and oat (Harrison and Kaufmann, 1980). N-[2-isopentenyl] adenine (2iP) promotes tillering in rice (Suge and Iwamura, 1993) as does the foliar application of the antiauxin 2,3,5,-triiodobenzoic acid (TIBA) (Yamada *et al*., 1963). However, these experiments were done in the laboratory using tiller bud segments as materials (Langer *et al*., 1973; Harrison and Kaufmann, 1980; Suge and

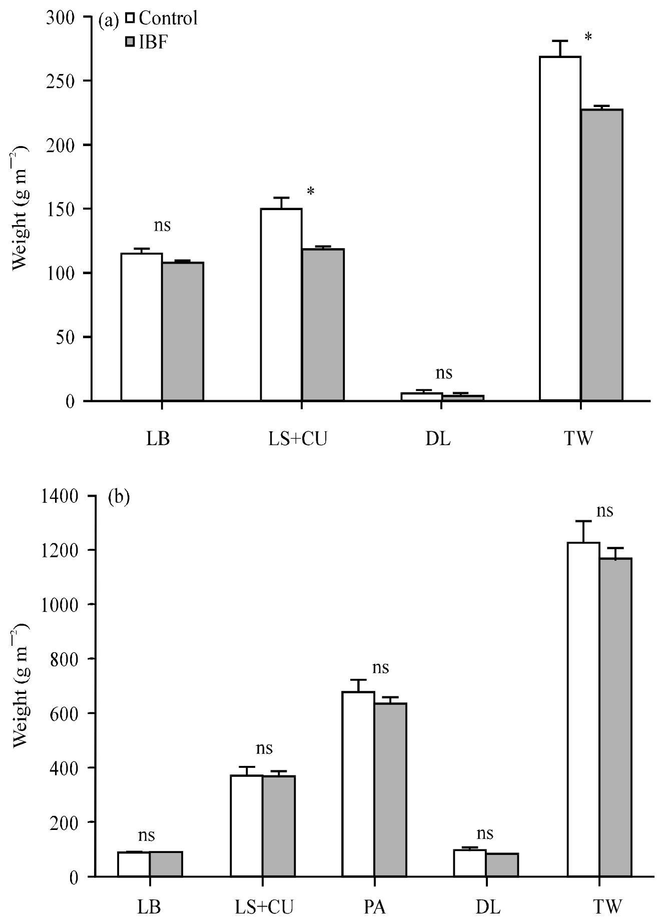

Fig. 6(a-b): Changes in dry weight of each organ of rice in the presence or absence of IBF at different growth stages at (a) Maximum tiller number stage and (b) Harvest time. LB: Leaf blade, LS: Leaf sheath, CU: Culm, DL: Dead leaf, PA: Panicle, TW: Total weight. Bars show means±SE (n = 3). *: Significant difference compared to the control at the 5% level, ns: No statistical difference between the control and IBF treatments

Table 1: Comparison of grain yield and yield components of rice between the IBF-treated and control plants

Treatment	No.of panicles per hill	No. of grains per panicle	Ripened grain (%)	Thousand grain wt. (g)	Grain yield (g m^{-2})
Control	432.8±12.6	63.3±2.2	87.0±1.8	22.8±0.1	523.9±19.3
IBF	538.9±21.0*	47.8±1.2**	90.4±0.6ns	22.9±0.5ns	497.0±17.3ns

Means±SE (n = 3) for each treatment are shown, *,**: Significant difference compared the control at 5 and 1% level, respectively. ns: No difference between control and IBF treatments

Iwamura, 1993) or carried out in potted conditions (Yamada *et al.*, 1963). Since IBF treatment promoted tillering in field conditions in the present study, the practical use of IBF can be more easily applied to actual rice cultivation. The TIBA and 2iP treatments promoted tillering by altering the parallelism of leaf emergence between main stem and tiller and the tillering itself, including secondary or tertiary tillers, was hastened. This resulted in more tillers being produced at high leaf positions where the untreated control plants had not produced any tillers in rice (Yamada *et al.*, 1963) and oat (Suge and Iwamura, 1993). Although, IBF treatment may increase tillering in a similar manner to TIBA and 2iP, the mode of action of IBF on tillering in rice needs to be elucidated. These experiments show that the inhibitor of GA biosynthesis, IBF, is effective for promoting tillering in no-tillage rice cultivation with a single basal fertilization of CAF to the nursery box.

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