# 벼에 있어서 bensulfuron-methyl의 吸收 移行에 미치는 無機營養分과 混合除草劑의 影響

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Effect of Mineral Nutrients and Mixed Herbicides on the Absorption and Translocation of Bensulfuron-methyl in Rice

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#### **Abstract**

Absorption and translocation of bensulfuron-methyl {methyl 2[[[[[(4,6-dimethoxy-2-pyrimidinyl) amino]carbonyl]amino]sulfonyl]methyl]benzoate} in rice (*Oryza sativa* L.) as affected by mineral nutrients and mixed herbicides were determined using the <sup>14</sup>C-labeled herbicide in culture solution. Absorption of <sup>14</sup>C-bensulfuron-methyl by the root decreased with increasing concentration of bensulfuron-methyl. However, increase in the application concentration did not affect movement of the <sup>14</sup>C to the shoot. There was no difference in total amount of <sup>14</sup>C-bensulfuron-methyl taken up between absorption periods of 12 and 48 hours, whereas <sup>14</sup>C-bensulfuron-methyl translocated to the shoot increased with increasing the absorption period. When bensulfuron-methyl mixtures were applied, butachlor [N-(butoxymethyl)-2-chloro-N-(2',6'-diethylphenyl)acetamide] did not affect absorption and translocation of <sup>14</sup>C-bensulfuron-methyl. However, quinclorac (3,7-dichloro-8-quino-line carboxylic acid) mixed at a high concentration resulted in decrease in absorption and translocation of <sup>14</sup>C-bensulfuron-methyl. Nutritional disorder such as deficient or excess supply of mineral nutrients caused to inhibit absorption of <sup>14</sup>C-bensulfuron-methyl. The greatest decrease and delay of <sup>14</sup>C-bensulfuron-methyl absorption and/or translocation occurred in N-deficient and S-excess supply conditions.

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#### INTRODUCTION

Bensulfuron-methyl, a sulfonylurea herbicide, is widely used in transplanted rice to selectively control annual and perennial broadleaf weeds and sedges<sup>1)</sup>. To increase the spectrum of weed control, however, bensulfuron-methyl is applied in combination with a 'grass' effective herbicide such as butachlor or quinclorac. When the mixtures are used as pre-emergence treatment, an adverse effect may occur in rice plants aside from an improvement in weeding effect.

Interactions of a herbicide with other herbicides or fertilizers have long been recognized and determined by several workers<sup>2,3)</sup>. Hatzios and Penner<sup>3)</sup> indicated that interactions between agrochemicals may result in adverse effects and these may be due either to alterations of the absorption, translocation, and biotransformation of one component from another, or to combinations of their actions or effects on target or nontarget species. Enhanced uptake and/or transport of picloram (4-amino-3,5,6-trichloro-2-pyridinecarboxylic acid) is evident in the presence of phenoxyalkanoic acid herbicides <sup>4,5)</sup>. Addition of ammonium nitrate or calcium chloride also results in increases in herbicide penertration and translocation<sup>6,7)</sup>.

The objective of this research was to investigate the absorption and translocation patterns of bensulfuron-methyl in rice as affected by mineral nutrients and the mixed herbicides. Experiments were carried out to: a) determine the effect of application time and concentration of bensulfuron-methyl. b) evaluate the effect of butachlor and/or quinclorac mixtures, and c) observe the effect of deficiency or excess supply of major mineral nutrients.

#### MATERIALS AND METHODS

Growth of rice plants Rice (cv. Choochung) seeds were selected, sterilized, and soaked in distilled water prior to pre-germination. When the coleoptile reached 5mm long, ten uniform seedlings were selected and transferred to a plastic pot (15×20cm) containing washed sands and culture solution8, and raised by 2-leaf stage in a greenhouse where the temperature varied between 21 and 30°C and natural daylight provided approximately 270µE·m<sup>-2</sup>·s<sup>-1</sup> photosynthetically active radiation during the experimental period. The seedlings were subjected to growing under different conditions of the culture solutions; fullstrength, nutrient deficiency, and excess supply of nutrients. Nutrient deficient condition was prepared by deleting respective major nutrients from full-strength solution, whereas for condition of excess supply of nutrients the respective major nutrients were added two-fold as much as required by full-strength solution. The nutrient solution was replenished daily. At 2-leaf stage of the seedlings all the culture solutions were changed with the corresponding fresh solutions and the seedlings were grown for another three days prior to application of herbicides. There were four replications.

<sup>14</sup>C-treatment solution <sup>14</sup>C-bensulfuron-methyl, uniformly labeled at the phenyl ring and having a specific activity of  $14.9\mu$ Ci/mg, was used. After <sup>14</sup>C-bensulfuron-methyl was dissolved in acetonitrile, the solution was re-diluted in deionized water. Five ml of the labeled herbicide and the required volume of unlabeled bensulfuron-methyl prepared from 93% technical grade were added to the nutrient solution to have 0.05 μCi

per pot. The inclusion of unlabeled bensulfuronmethyl was to adjust herbicide concentrations. The final herbicide concentrations in the treatment solution were adjusted to 0.1, 1, 10, and 100  $\mu M$ .

**Experimental treatments** Three experimental conditions were employed: 1) The rice seedlings were treated with concentrations of bensulfuron-methyl ranging from 0.1 to 100µM. The plants were allowed to take up the nutrient solution over a period of 24 h and then harvested. To determine the effect of application time, bensulfuron-methyl at 10µM was applied to the plants for 12-, 24-, and 48-h uptake period. 2) Herbicide mixtures of bensulfuron-methyl used were butachlor and quinclorac. The respective herbicide at 0.1, 1, and 10µM was combined with 10µM bensulfuron-methyl. The plants were harvested after completion of the 24-h uptake period. 3) Nutritional stresses (deficiency and/or excess supply) subjected to the plants were for N, P, K, Ca, Mg, and S. bensulfuron-methyl at 10µM was treated to the plants for 12 and 48 h.

Plant harvest and <sup>14</sup>C assay After harvest, the plants were rinsed twice with deionized water to remove unabsorbed herbicide and divided into roots and shoots. The plant parts were dried at 80°C for 3 days and weighed. Two hundreds mg of the dried sample were combusted in a biological sample oxidizer. The liberated <sup>14</sup>CO<sub>2</sub> was collected in 7ml of a scintillant cocktail containing a CO<sub>2</sub> absorbent and radioassayed using a liquid scintillation spectrometry(Beckman LS 51 00).

#### **RESULTS AND DISCUSSION**

Absorption and translocation Uptake of

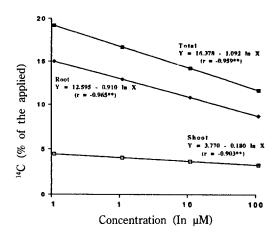


Fig. 1. Distribution of <sup>14</sup>C-bensulfuron-methyl as affected by the application concentrations.

Table 1. Absorption and translocation of <sup>14</sup>C-bensulfuron-methyl as affected by time of application

Time after application	Distribution of radioactivity (% of applied)			
(h)	Absorption	Root	Shoot	
12	10.8	9.2	1.6	
24	11.7	8.8	2.9	
48	11.7	8.2	3.5	
LSD (0.05)	1.0	0.6	0.3	

bensulfuron-metyhy was affected by concentration of the herbicide (Fig. 1). Total amount of <sup>14</sup>C absorbed by the root decreased with increasing the application concentration. After absorption, about two-thirds of <sup>14</sup>C was found in the roots and the rest in the shoots. The <sup>14</sup>C translocated to the shoot was not influenced by the application concentration, whereas the <sup>14</sup>C remained in the root was dependent upon the total <sup>14</sup>C absorbed. This result agrees with the findings of Yuyama *et al.*<sup>9</sup>, indicating a limited translocation of absorbed <sup>14</sup>C-bensulfuron-methyl from rice roots to shoots.

Root absorption of bensulfuron-methyl occurred

Mixed herbicide	Concentration (µM)	Distribution of radioactivity		
		(% of applied)*		Root/Shoot
		Root	Shoot	Ratio
Untreated	_	10.5ab	3.7ab	2.8
Butachlor	0.1	11.3a	3.8a	3.0
Butachlor	1	10.5ab	3.3b	3.2
Butachlor	10	10.2bc	3.6ab	2.8
Quinclorac	0.1	10.4b	3.7ab	2.8
Quinclorac	1	10.2b	2.8c	3.6
Quinclorac	10	9.0c	2.4c	3.8

Table 2. Effect of mixed herbicides on absorption and translocation of <sup>14</sup>C-bensulfuron-methyl in rice.

within a relatively short period of time (Table 1). There was no significant difference in <sup>14</sup>C absorbed between 12-h and 48-h absorption periods. However, the amount of <sup>14</sup>C-bensulfuron-methyl translocated to the shoots increased as the exposure time increased, resulting in decrease in <sup>14</sup>C of the roots. Yuyama *et al.*<sup>9)</sup> reported that the uptake of bensulfuron-methyl in paddy rice rapidly increased by 6 h and thereafter showed a gradual increase by 24h.

The regression analysis of the data showed a negative linear relationship between absorption of bensulfuron-methyl and concentration of bensulfuron-methyl in the ambient solution (r=-0.959, p<0.01). This negative linearity indicated that the mechanism of absorption may not be based on a simple diffusion process against a concentration gradient. Devine *et al.*<sup>10)</sup> determined that chlorsulfuron [2-chloro-N-[[4-methoxy-6-methyl-1,3,5-triazin-2-yl) amino] amino] carbonyl] benzenesulfonamide} uptake into excised pea root tissue over a concentration range from 0.001 to 1 mM was linear and suggested that the absorption mechanism was a non-facilitated process. The difference may be due either to different absorption process

itself between the herbicides or to difference in plant materials used. In this experiment intact plants were used.

Effect of mixed herbicides Absorption and translocation of <sup>14</sup>C-bensulfuron-methyl varied with the kind of herbicides combined (Table 2). Butachlor at the application concentrations ranging from 0.1 to 10 µM did not greatly affect the total amount of <sup>14</sup>C absorption, whereas a significant decrease in absorption of <sup>14</sup>C-bensulfuron-methyl was found when 10 µM quinclorac was mixed.

The pattern of translocation of <sup>14</sup>C-bensulfuron-methyl was related with amount of <sup>14</sup>C absorbed. Butachlor mixture did not alter the root/shoot ratio as compared with the untreated control. However, increase in the ratio occurred when higher than 1 µM of quinclorac was applied, resulting in decrease in translocation of <sup>14</sup>C-bensulfuron-methyl absorbed. This indicated that quinclorac caused to decrease both absorption and translocation of bensulfuron-methyl.

Based on the results obtained, it is assumed that both the herbicide mixtures employed do not affect membrane integrity of the root cells. If the

<sup>\*</sup> Means in a column followed by a common letter are not significantly different at 5% level by Duncan's Multiple Range Test.

membranes disrupted mechanically by the herbicide mixtures, amounts of <sup>14</sup>C absorbed and translocated would have rather been increased as compared with the untreated control. Instead the decreased absorption and translocation of <sup>14</sup>C-bensulfuron-methyl by means of quinclorac may be due to either to competition for site of uptake or to interfering result on absorption process of bensulfuron-methyl.

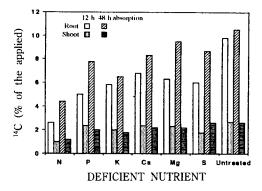


Fig. 2. Effect of deficiency of mineral nutrients on distribution of <sup>14</sup>C-bensulfuronmethyl after 12-h and 48-h absorption periods.

Effect of mineral nutrients Absorption and translocation of 14C-bensulfuron-methyl in the rice plants was affected by mineral nutrients (Fig. 2, 3). When the rice plant was grown under deficient conditions of mineral nutrients, the absorption of 14C decreased and delayed. The greatest decrease in the absorption occurred in N-deficiency, but the least in Mg-deficiency (Fig. 2). In addition, the absorption process tended to be delayed. Unlike a rapid absorption of 14C-bensulfuron-methyl in the untreated control, deficient mineral nutrition resulted in continuous absorption of <sup>14</sup>C by 48-h absorption period. However, the translocation of the 14C absorbed was not much affected by the mineral nutrition, except for N-

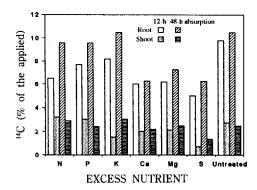


Fig. 3. Effect of excess supply of mineral nutrients on distribution of <sup>14</sup>C-bensulfuron-methyl after 12-h and 48-h absorption periods.

deficiency. The <sup>14</sup>C translocated under N-deficient condition was reduced as compared with the untreated control.

Excess supply of N, P, and K caused a delayed absorption of <sup>14</sup>C-bensulfuron-methyl, but did not affect the total amount of the herbicide absorbed (Fig. 3). In contrast, excess supply of Ca, Mg, and S resulted in reduction of the absorption of <sup>14</sup>C-bensulfuron-methyl. Translocation of the absorbed <sup>14</sup>C was not affected by excess supply of the mineral nutrients, except for S. Decrease in the <sup>14</sup>C translocated was found in S excess supply.

These results indicate that absorption and translocation of bensulfuron-methyl is influenced by the application concentration, kind of mixed herbicides, and mineral nutrients of rice. Whatever the factors employed are involved, amount of <sup>14</sup>C-bensulfuron-methyl absorption and translocation tends to decrease. This suggests that phytotoxic mode of action of bensulfuron-methyl in rice is not associated with the total amount of the herbicide absorbed and translocated. Pyon and Kwon<sup>110</sup> reported that differential selectivity to the herbicide between rice and *Cyperus serotinus* was due

### 要 約

벼에 있어서 bensulfuron-methyl의 흡수 이행에 미치는 무기영양성분 및 혼합제초제의 영향을 수경 액상에서 <sup>14</sup>C-除草劑로 조사하였다. 뿌리에 의한 <sup>14</sup> C-bensulfuron-methyl의 吸收는 處理濃度의 증가에 따라 감소되었지만, 농도 증가가 줄기로의 移行에는 영향을 미치지 않았다. 14C-bensulfuron-methyl의 總 吸收量은 吸收 12시간 및 48시간 사이에 차이를 보 이지 않았던 반면에, 줄기로의 移行은 吸收時間이 길수록 증가되었다. 14C-bensulfuron-methyl의 吸收 移行은 混合除草劑 butachlor에 의하여 영향을 받지 않지만, quinclorac이 高濃度로 혼합될 때 14C-bensulfuron-methyl의 흡수 이행이 감소되었다. 무기성 분의 缺乏 및 過量供給은 <sup>14</sup>C-bensulfuron-methyl의 홈수를 沮害하였다. <sup>14</sup>C-bensulfuron-methyl의 흡수 이행은 窒素의 결핍 및 黃이 과량 공급될 때 가장 심하게 감소되고 지연되었다.

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