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The Mechanism of Bentazon Selectivity

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Absorption, translocation and metabolism of [^{14}C]3-isopropyl-2,1,3-benzothiadiazin-4-one-2,2-dioxide (bentazon) by several plant species were investigated to determine the mechanism of bentazon selectivity.

Marked selective phytotoxicities were observed between resistant rice (*Oryza sativa* L.) and susceptible *Cyperus serotinus* Rottb. when treated with bentazon. Absorption and transolcation of bentazon did not differ greatly between highly resistant rice and susceptible *C. serotinus*. However, a marked difference in bentazon metabolism occurred between the two species. In rice about 80% of the absorbed bentazon was metabolized within 24 h, and after 7 days about 85% was converted to a major water-soluble metabolite and unchanged bentazon was only 5%. In *C. serotinus* 50–75% of the radioactivity was unchanged bentazon after 7 days.

Large amounts of water-soluble metabolites were detected in root-treated resistant plants such as barnyardgrass (*Echinochloa crus-galli* Beauv.), soybean (*Glycine max* Merr.) and corn (*Zea mays* L.), but only small amounts were present in such susceptible plants as *Sagittaria pygmaea* Miq. and *Eleocharis kuroguwai* Ohwi. Therefore, the mechanism of bentazon selectivity appears to be a difference between resistant and susceptible species in their ability to metabolize and detoxify bentazon.

The major metabolite in rice was identified as 6-(3-isopropyl-2,1,3-benzothiadiazin-4-one-2,2-dioxide)-*O*-β-glucopyranoside, determined by GC-MS, NMR, IR and gas chromatography after hydrolysis with sulfuric acid or β-glucosidase.

INTRODUCTION

Bentazon, 3-isopropyl-2,1,3-benzothiadiazin-4-one-2,2-dioxide is a selective herbicide for weed control in soybean (1, 2), cereals (3, 4) and rice (5, 6, 7). Bentazon is generally used as a postemergence herbicide by foliar application. In rice culture bentazon is effective by both foliar and flooded-water application under flooded rice field conditions.

Marked selective phytotoxicities were observed between resistant and susceptible plants when treated with bentazon. Mine *et al.* (6) investigated the herbicidal effect on 43 species of paddy weeds and reported that a marked selectivity between the plants was observed. Many graminaceous plants such as rice (*Oryza sativa* L.), corn (*Zea mays* L.), barley (*Hordeum vulgare* L.), barnyardgrass (*Echinochloa crus-galli* Beauv.) and most grassy weeds, and soybean (*Glycine max* Merr.) are resistant. Bentazon susceptible plants are broadleaved weeds and sedges such as pigweed (*Amaranthus spp.*), common lamb's-quarters (*Chenopodium album* L.), common cocklebur (*Xanthium pensylvanicum* Wallr.), smartweed (*Polygonum pensylvanicum* L.), false pimpernel (*Lindernia pyxidaria* L.), *Sagittaria pygmaea* Miq., *Eleocharis kuroguwai* Ohwi., purple nut-

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sedge (*Cyperus rotundus* L.) and *Cyperus serotinus* Rottb.

Mine and Matsunaka (8, 9) have reported that photosynthetic inhibition may be the primary mode of action of bentazon under flooded rice field conditions. Severe inhibition of CO_2 fixation was observed in both rice and *Cyperus serotinus* plants soon after application of bentazon. However, in resistant rice almost total recovery of photosynthetic inhibition was observed within 5 days, but no recovery occurred in susceptible *C. serotinus*. The authors postulated that a detoxication of bentazon may occur in resistant rice plant (8, 9). Herbicidal selectivity may be primarily due to differential metabolism and detoxication of herbicides in plants. This basis for selectivity has been demonstrated for simazine (10, 11), atrazine (12), propanil (13, 14), amiben (15), and pyrazone (16).

Zaunbrecher and Rogers (17) reported no differences in the absorption and translocation of bentazon by susceptible cocklebur and resistant soybean after foliar application. Hayes and Wax (18) reported no significant differences in absorption of bentazon by leaf disks of between tolerant and sensitive soybean cultivars over a 10-h incubation period. They also reported only slight differences in translocation and metabolism of bentazon between tolerant and sensitive cultivars of soybean by foliar application (18). Otto and Dresher (19) found water-soluble metabolites of [^{14}C]bentazon in foliar and root-treated soybean after 50 and 28 days, respectively.

This investigation was undertaken to determine the mechanism of bentazon selectivity in plants. Highly resistant rice plants were compared to susceptible *C. serotinus* plants with respect to absorption, translocation, and metabolism of bentazon. In addition, metabolism of bentazon was determined in other resistant and susceptible crop plants and weeds.

MATERIALS AND METHODS

Plant materials. Rice (*Oryza sativa* L. cultivar Nihonbare), corn (*Zea mays* L., cultivar Koushu), soybean (*Glycine max* Merr., cultivar Okuharawase), *Cyperus serotinus* Rottb., barnyardgrass (*Echinochloa crus-galli* Beauv. var. *oryzicola* Ohwi), *Sagittaria pygmaea* Miq. and *Eleocharis kuroguwai* Ohwi were cultivated in sand in a glasshouse at 23–28°C. Plants were fertilized with Kimura's B nutrient solution (8, 20). Before treatment with bentazon, plants grown in sand were removed and transferred to flasks containing 100 ml of the nutrient solution unless otherwise described. The flasks were covered with aluminium foil to shut out light.

Chemicals. The 50% wettable powder formulation of bentazon was used to investigate the selective herbicidal effects. Labeled [^{14}C]10-bentazon (specific activity, 2.99 mCi/m*M* was used for investigating absorption, translocation and metabolism in plants. For the identification of the major metabolite in rice, purified bentazon (99%) was used.

Selective herbicidal effect between rice and C. serotinus. Rice and *C. serotinus* plants, grown in solution culture to the five- to six-leaf stage, were treated with bentazon in 1.2 liter bottles. The roots of the plants were treated with the nutrient solution containing 1 liter of 0.8–200 ppm bentazon. The plants were placed in a glass chamber placed outdoors at 23–28°C and approximately 70–80% relative humidity. The plants were harvested and their fresh weights determined after 20 days. The experiments were conducted in duplicate and repeated twice.

Absorption, translocation, and metabolism in plants. Rice and *C. serotinus* plants at the four- to five-leaf stage of growth were root-treated with 3 ppm [^{14}C]bentazon in 100 ml of nutrient solution or treated with 0.06 μCi of the radioactive herbicide on the middle of the second leaf from the top. The treated plants were placed in a growth chamber with a 12-h photoperiod, 28 ± 2°C day temperature and 23 ± 2°C night temperature, 60–70% relative humidity and light intensity of 5000–7000 lux. Absorp-

tion, translocation and metabolism of bentazon were determined periodically after treatment. The treated parts of the plants were rinsed once in 20% acetone (v/v) and then in distilled water. Each plant part was cut with a pair of scissors and homogenized and extracted with cold 80% acetone (v/v) with a mortar and pestle for three times.

The crude 80% acetone extracts from the root-treated plants and culture solution were radioassayed at 1, 2, 4 and 7 days with liquid scintillation spectrometer (Beckman, LS-100) for determing the absorption through the plants. The ^{14}C activity remaining in the 80% acetone-insoluble plant residue was radioassayed after dry combustion in oxygen with an autosample-oxydizer (Packard, Tricarb 305).

To examine the translocation through the plants, crude extracts from the leaf, sheath, stem and root of the plants root-treated with bentazon for 5 days were radioassayed.

For comparing the metabolism pattern of the plant species by root or foliar application, the crude extracts evaporated to dryness were developed on a silica gel TLC (Merck, F-254, 0.25 mm thick) with $CHCl_3$: MeOH, 7:3 (v/v). Radioactive zones were detected with a radiochromatogram scanner (Aloka, TRM-1B).

For qualitative and quantitative assay, aqueous solution (pH 7.0) of the crude extracts was partitioned with benzene and then acidified with 1 *N* HCl to pH 1.5 and was again partitioned with benzene for three times each. The aqueous layer was neutralized by adding 1 *N* NaOH. Aliquots from each aqueous and total benzene layer were developed on a silica gel TLC with ethyl acetate: isopropanol:water, 5:2:0.5 (v/v/v). The gel from the radioactive zones detected by radioautography with X-ray film were scraped and radioassayed with a liquid scintillation spectrometer. The nutrient solution was also developed on a silica gel TLC and radioactive zones were assayed in the same way.

Metabolism of bentazon in various plant species. Rice, barnyardgrass, corn, *Cyperus serotinus*, and *Eleocharis kuroguwai* in their four- to five-leaf stage, soybean in the three- to four-leaf stage and *Sagittaria pygmaea* in the six- to eight-leaf stage were root-treated for 4 days with 3 ppm [^{14}C]bentazon in the nutrient solution. Crude 80% acetone extracts from the treated plant leaves were partitioned, chromatographed and radioassayed in the same way as the previous experiments.

Isolation of the major metabolite in rice. The fresh root and sheath of rice plant (230 g) root-treated with 350 mg bentazon in a nutrient solution for 7 days were extracted with MeOH (900 ml × 3) and yielded of crude syrup (8.1 g). The crude syrup was washed with *n*-hexane (20 ml × 3) and $CHCl_3$ (20 ml × 3) and the residual part which solubilzed to MeOH was chromatographed over silica gel tlc (Merck, F-254, 0.5 mm thick, developed with $CHCl_3$: MeOH, 1:1, v/v). The UV light absorptive part (R_f 0.45–0.50) was collected and eluted with MeOH yielded 126 mg of glassy material (I). NMR(acetone-d_6): δ7.73 (1H, d, J = 3 Hz for H-5), 7.43 (1H, dd, J = 8.5, 3 Hz for H-7), 7.17 (1H, d, J = 8.5 Hz for H-8), 5.01 (1H, d, J = 7.0 Hz for C_1'–H), 5.3–4.65 (1H, m. for isopropylmethine), 4.5–3.3 (*ca.* 11H for glucose-H), 1.52 (6H, d, J = 6.5 Hz for ispropyl-CH_3 × 2).

Enzymatic hydrolysis of (I) and identification of the hydrolyzate (II). A solution of 50 mg of (I) in 10 ml of 0.01 *M* NaOAc buffered with HOAc to pH 5.0 was added to 20 mg of emulsin (β-glucosidase)[3]; the resultant solution was allowed to shake at 35°C for 2 hr. The mixture was extracted with 10 ml of EtOAc twice and the extract was dried with Na_2SO_4. The EtOAc was evaporated and yielded 21 mg of yellowish syrup. The syrup was purified over a silica gel tlc (developed with $CHCl_3$: MeOH, 7:3, v/v, R_f 0.55) and obtained as a crys-

[3] Purchased from Miles Laboratories Ltd., England.

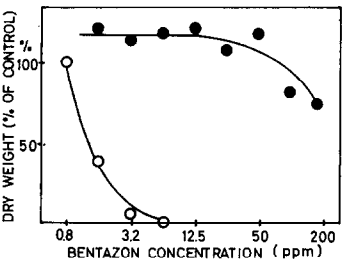


FIG. 1. *Selective herbicidal effect of bentazon on rice and* Cyperus serotinus. *Rice and* C. serotinus *at the five- to six-leaf stage were root-treated with bentazon in a nutrient solution. The plants were grown in a glass chamber at 23–28°C for 20 days. rice* (●), Cyperus serotinus (○).

talline (II), which was purified by recrystallization from EtOAc giving the 6-OH bentazon as needles (16 mg): GC-MS; M^+ 256, 241, 214, 198, 177, 150, 135, 107, 80. The material was identical with authentic 6-OH bentazon[4] by NMR an IR.

Acid hydrolysis of (I) and identification of sugar moiety. A solution of 15 mg of (I) in 4 ml of MeOH was mixed with 4 ml of 0.1 *N* H_2SO_4; the resultant solution was kept at 90°C for 1 h. The solution was concentrated *in vacuo* to *ca.* 3 ml and washed with EtOAc (3 ml × 3). The aqueous layer was neutralized with $NaHCO_3$ and filtered. The aqueous filtrate was concentrated to dryness yield a thick syrup which was extracted with MeOH (10 ml × 2). The MeOH extract yield a clear syrup (5 mg). To a solution of the syrup in 0.5 ml of pyridine (dry) was added 0.5 ml of $(Me)_3SiCl$ and 0.5 ml of $[(Me)_3Si]_2NH$. After 5 min, the solution was evaporated to dryness. The residue was dissolved in dry CCl_4 and filtered. The filtrate was gas chromatographed and found to contain only 1,2,3,4,6-penta-*O*-trimethylsilyl-D-glucopyranose by co-gas chromatographic analysis with an authentic sample. Gas chromatographic conditions: Shimadzu GC-3BF, FID; (5% SE-30, glass column 3 mm × 1 m, 170°C, carrier N_2 48 ml/min) t_R, α-epimer 6′45″, β-epimer 10′10″; (5% LAC-2R-446, glass column 3 mm × 1.7 m, 120°C, carrier N_2 36 ml/min) t_R, α-epimer 3′55″, β-epimer 7′10″.

Herbicidal activity of the major metabolite (I) in rice. Sagittaria pygmaea plants at the five- to six-leaf stage were root-treated in 100 ml of nutrient solution with 1–100 ppm of bentazon or the major metabolite (I) in rice. The treated plants were placed in a glass chamber at 23–28°C and their fresh weight of leaves was determined after 14 days.

RESULTS AND DISCUSSION

The basis for selectivity was investigated between resistant rice and susceptible *Cyperus serotinus* (Fig. 1). *C. serotinus* plants were almost completely killed by root treatment with 3.2 ppm of bentazon in the nutrient solution. However, rice plants were extremely resistant to bentazon and were not injured at the concentration of less than 50 ppm. Rice plants were not killed even at 200 ppm of bentazon. The results indicated that rice plants were able to tolerate at least a 100-fold increase in bentazon concentration as compared to *C. serotinus*. The growth of rice plants in the control plots was somewhat retarded because of the competitive effects of *C. serotinus* in the same pots, and dry weight of the rice plants treated with lower doses of bentazon was over 100% as compared to that of control plots. A marked selectivity between rice and *C. serotinus* was also observed when plants were treated by foliar application (6).

The absorption, translocation, and metabolism of bentazon in resistant rice and susceptible *C. serotinus* plants were investigated to determine the basis for selectivity. Absorption of bentazon through root by both species is shown in Table 1. The amount of absorbed ^{14}C activity in the plants was even higher in resistant rice than in susceptible *C. serotinus*. Within 24 h, the concentrations of absorbed ^{14}C activity in the plants were 526 and 195 dpm/mg,f.w. in rice and *C. serotinus*, re-

[4] Authentic sample of 6-OH bentazon was afforded from BASF Company, Germany, F.R.

TABLE 1

Absorption of [^{14}C]Bentazon in Rice and Cyperus serotinus *Plants after Root-Uptake from 3 ppm Aqueous Solution*

Plant species	Days after treatment	Nutrient solution ^{14}C-activity		Whole plant ^{14}C-activity		Recovery	Concentration in whole plant (dpm/mg, f.w.)
		$\times 10^4$ dpm	%	$\times 10^4$ dpm	%	%	
Rice	1	647.7	77.8	120.7	14.5	92.3	526
	2	462.9	55.6	281.4	33.8	89.4	974
	4	345.5	41.5	388.8	46.7	88.2	1146
	7	136.5	16.4	586.1	70.4	86.8	1519
Cyperus serotinus	1	711.8	85.5	74.9	9.0	94.5	195
	2	603.6	72.5	152.3	18.3	90.8	277
	4	492.0	59.1	267.2	32.1	91.2	638
	7	381.3	45.8	365.5	43.9	89.7	810

spectively. The concentrations of absorbed radioactivity in the whole plant remained higher in rice than in *C. serotinus* up to 7 days after treatment. Susceptible *C. serotinus* plants were killed beyond the 7-day period.

Distribution of 80% acetone extracted ^{14}C activity in the plants root-treated with [^{14}C]bentazon for 5 days is shown in Table 2. ^{14}C activity was distributed in all plant parts of both species. These results indicate that bentazon was absorbed by roots and readily translocated to stems, sheaths, and leaves in both species. Rice plants showed a high concentration of ^{14}C activity in the stem. The concentration of ^{14}C activity in the leaf was very similar between rice and *C. serotinus*. The results indicate that differences in absorption and translocation of bentazon by resistant rice and susceptible *C. serotinus* may not be responsible for the selective action of bentazon between the two species.

TABLE 2

Distribution of ^{14}C Activity in Rice and Cyperus serotinus *Plants after 5 Days from Root-Treatment of [^{14}C]Bentazon*

	Rice		*Cyperus serotinus*	
	Extracted activity ($\times 10^4$ dpm)	Concentration in plant (dpm/mg, f.w.)	Extracted activity ($\times 10^4$ dpm)	Concentration in plant (dpm/mg, f.w.)
Leaf	2.1	250	7.0	320
Sheath	3.4	550	7.6	250
Stem	3.5	3890	3.1	650
Root	7.0	1460	10.0	950

The metabolism of bentazon in plants was investigated as a basis for selectivity between rice and *C. serotinus*. Crude 80% acetone extracts from [^{14}C]bentazon-treated plants were chromatographed on thin-layer plates with a solvent system, $CHCl_3$: CH_3OH, 7:3. The distribution of the radioactivity on the plates is shown in Fig. 2. The extract from rice indicated a high concentration of metabolite at R_f 0.17 as compared to unchanged bentazon (R_f 0.48). This was true of both root and foliarly applied [^{14}C]bentazon. The rate of [^{14}C]-bentazon metabolism in rice appeared to be higher in roots than in leaves. In *C. serotinus*, most of the radioactivity was detected as unchanged bentazon. These results suggested that rice metabolized bentazon much faster than *C. serotinus*.

For quantitative assay, bentazon metabolism in plants was determined after 1, 2, 4 and 7 days from root treatment with [^{14}C]bentazon (Tables 3, 4). After partitioning of the crude extracts, bentazon (R_f 0.59) and a few metabolites (R_f 0.24,

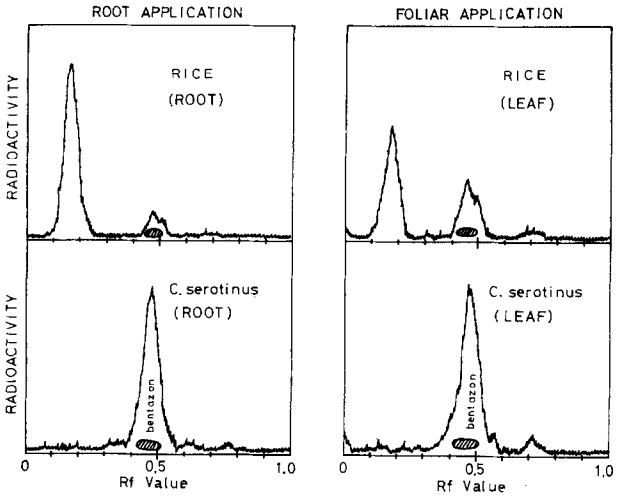


FIG. 2. *Radioactive compounds extracted from roots and leaves of rice and* Cyperus serotinus *treated with [^{14}C]bentazon for 4 days. The bentazon-treated plants were extracted with 80% acetone and chromatographed on a silica gel thin-layer plate, and radioactivity was detected with a radiochromatogram scanner.*

0.68, 0.72) were detected in benzene layer by silica gel tlc developed with ethyl acetate: isopropanol:water (5:2:0.5, v/v/v). A major metabolite in rice (R_f 0.24) was predominantly in water layer. Little decomposition of [^{14}C]bentazon was observed in the nutrient solution used for the culture of rice and *C. serotinus*. In rice (Table 3), metabolism of absorbed bentazon occurred greatly and percentage of the metabolites in shoot and root was almost same. Eighty to eighty-five percent of the extracted radioactivity was that of the metabolites within 24 h and unchanged bentazon was only about 5% after 7 days. About 80–85% of the absorbed bentazon was converted to a major water-soluble metabolite after 7 days. In *C. serotinus* (Table 4), the metabo-

TABLE 3

Metabolism of [^{14}C]Bentazon in Rice Plant after Root-Uptake from 3 ppm Aqueous Solution

Plant part	Days after treatment	Concentration of absorbed ^{14}C activity (dpm/mg,f.w.)	80% Acetone extracted			Plant residue
			Benzene layer		Water layer	
			Bentazon (%)	Metabolites (%)	metabolites (%)	(%)
Shoot	1	459.2	15.9	9.3	69.5	5.3
	2	904.6	7.6	5.5	80.7	6.2
	4	1138.3	7.2	2.2	85.1	5.5
	7	1614.3	6.1	2.4	85.8	5.7
Root	1	791.8	12.9	7.9	72.0	7.2
	2	1237.2	3.8	3.4	83.5	9.3
	4	1170.6	3.9	1.5	84.5	10.1
	7	1241.0	4.1	1.8	86.1	8.0

TABLE 4

Metabolism of [^{14}C]*Bentazon in* Cyperus serotinus *Plant after Root-Uptake from 3 ppm Aqueous Solution*

Plant part	Days after treatment	Concentration of absorbed ^{14}C activity (dpm/mg,f.w.)	80% Acetone extracted			Plant residue (%)
			Benzene layer		Water layer	
			Bentazon (%)	Metabolites (%)	metabolites (%)	
Shoot	1	127.6	82.6	2.4	5.8	9.2
	2	229.6	81.7	2.4	6.6	9.3
	4	603.3	76.9	4.5	12.5	6.1
	7	797.4	75.4	2.9	16.1	5.6
Root	1	577.1	64.4	8.7	8.6	18.3
	2	522.6	59.7	7.4	9.9	23.2
	4	838.5	53.7	6.0	15.0	25.3
	7	879.5	51.4	3.4	20.6	24.6

lism of bentazon in root was greater than in shoot, but much less than in rice plants. The metabolites accounted for only 20–30% of the extracted radioactivity after 7 days. Thus, the rapid metabolism of bentazon in rice decreased the concetration of the active herbicide within the plant. The relative concentrations of [^{14}C]bentazon in the shoot after 7 days were 74 and 509 dpm/mg,f.w. (2.7 and 18.3 ppm in fresh weight) in rice and *C. serotinus*, respectively.

The authors (8) reported that severe inhibition of CO_2 fixation was observed in both rice and *C. serotinus* soon after foliar application of bentazon. However, in resistant rice almost total recovery of photosynthetic inhibition was observed within 5 days, but no recovery occurred in susceptible *C. serotinus*. Photosynthetic inhibition may be the primary mode of action of bentazon (8). The inhibition and recovery of photosynthesis in rice and *C. serotinus* corresponded with the rate of bentazon metabolism in these species. Therefore, the mechanism of bentazon selectivity between rice and *C. serotinus*

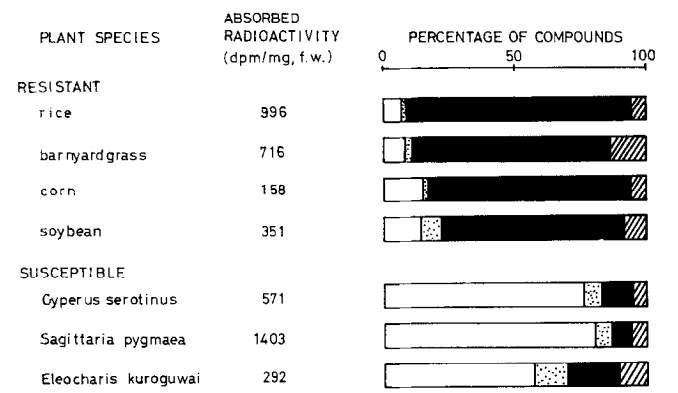
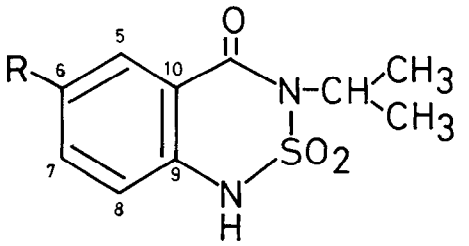


FIG. 3. *Metabolism of bentazon in various resistant and susceptible plants. Plants at the three- to eight-leaf stage of growth were root-treated with 3ppm of* [^{14}C]*bentazon in the nutrient solution for 4 days in a growth chamber at 23–28°C. Crude 80% acetone extracts from leaves of the treated plants were partitioned with benzene and water, and each layer was chromatographed on silica gel thin-layer plates. The radioactivity was assayed after extraction from radioactive zones on the plates.*

□ *Bentazon,* ▨ *metabolites in benzene layer* ■ *metabolites in water layer,* ▨ *plant residue.*



(I) R=O-β-D-glucopyranose

(II) R=OH

(III) R=H (bentazon)

FIG. 4. *Major metabolite in rice treated with bentazon.*

appeared to be a difference between the species in their ability to metabolize bentazon.

Similar metabolism experiments were conducted with rice and other resistant species such as corn, soybean and barnyardgrass, and susceptible species such as *C. serotinus, Sagittaria pygmaea,* and *Eleocharis Kuroguwai* (Fig. 3). All plants were root-treated with a solution of [^{14}C]bentazon for 4 days. The concentration of absorbed ^{14}C activity in the leaf was different among the plant species, and there was no correlation between the concentration and susceptibility to bentazon. Large amounts of a water-soluble metabolite (R_f 0.24–0.25) similar to that in rice was detected in resistant rice, corn, soybean and barnyard grass, but only small amounts of water-soluble metabolites were detected in susceptible *C. serotinus, S. pygmaea* and *E. kuroguwai.* In soybean, the two different water-soluble metabolites (R_f 0.17 and 0.25) were detected. The rate of bentazon metabolism in different plant species reflected their susceptibility to bentazon. These results confirmed the conclusion that the mechanism of bentazon selectivity appears to be a difference between resistant and susceptible species in their ability to metabolize and detoxify bentazon.

The major metabolite of bentazon in rice was determined. Methanol extracts of bentazon-treated rice afforded in about 40% yield a metabolite. The evidence presented below established that the compound is 6-(3-isopropyl-2,1,3-benzothiadiazin-4-one-2,2-dioxide)-*O*-β-D-glucopyranoside (I).

The NMR spectrum of (I) indicated that the sugar was present as a pyranoside and the benzene ring moiety exhibited eight isolated and well-resolved signals (in acetone-d_6, δ7.73 (1H, d, J = 3 Hz), 7.43 (1H, dd, J = 8.5, 3 Hz) and 7.17 (1H, d, J = 8.5 Hz). This splitting pattern of benzene ring has expected that the sugar group is at position 6 in comparison with the similar benzene splittings of 1,2-dimethyl-6-methoxy-4-quinoline (21) and 2,3-dihydro-6-methoxy-4-quinoline (22) (5, 7 and 8-methoxy analogues give different splittings).

β-Glucosidase hydrolysis of (I) afforded the authentic 6-OH bentazon (II), indicating a C_1'-β anomeric configuration and this result was consistent with the large (7.0 cps) coupling of the C_1' anomeric proton signal. The sugar obtained by acid hydrolysis of the metabolite (I) was shown to be D-glucopyranose by gas chromatography of the trimethylsilyl ether. Therefore, the major metabolite is 6-(3-isopropyl-2,1,3-benzothiadiazin-4-one-2,2-dioxide)-*O*-β-D-glucopyranoside (I).[5]

The herbicidal activity of the major metabolite (I) in rice was determined by root-treatment in a nutrient solution. The activity of the metabolite (I) was weak and *Sagittaria pygmaea* plants were not injured even at the highest concentration of 100 ppm. The fresh weight was 104% of the nontreated control. However, the plants

[5] Recently the authors found that Otto, BASF, had reported at WSSA meeting in 1975 that the glycosides of 6- and 8-hydroxybentazon were formed in about equal ratios and amounts in soybeans, and that in most other higher plants such as wheat, rice, peanuts and the weeds *Senecio* and *Chenopodium* spp., the 6-hydroxybentazon conjugate is formed predominantly.

treated with 1 ppm of bentazon were severely injured and the fresh weight was 17% of the control. At more than 3 ppm, the plants were completely killed.

Thus, the resistant plants rapidly metabolize bentazon to the nonphytotoxic metabolite and decreased the concentration of the active herbicide in plant. Therefore, the main mechanism of bentazon selectivity appears to be a difference between the plant species in their ability to metabolize and detoxify bentazon.

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REFERENCES

1. M. Luib and J. C. van de Weerd, Trial results obtained in soya beans with 3-isopropyl-1H-2,1,3-benzothiadiazin-4(3H)-one-2,2-dioxide, *Proc. 11th Brit. Weed Control Conf.* p. 547 (1972).
2. B. Wuerzer, J. Thompson, and J. W. Daniel, Application rate and timing of BAS-3512-H in soybeans, *Proc. S. Weed Sci. Soc. Amer.* **25**, 108 (1972).
3. S. Behrendt and L. Sipos, Ein Thiadiazinderivat gegen breitblattige Unkrauter in Wintergetreide, *3. Symposium uber neue Herbizide, Paris* (1969).
4. B. H. Menck and S. Behrendt, Trial in cereals with bentazon (3-isopropyl-1H-2,1,3-benzothiadiazin-4(3H)-one-2,2-dioxide) in combination with hormones, *Proc. 11th Brit. Weed Control Conf.*, p. 666 (1972).
5. M. Luib, S. Behrendt, S. Haaksma, and B. G. M. Kamp, Trials with bentazon in rice, *Proc. 4th Asian-Pacific Weed Sci. Soc. Conf.*, p. 140 (1973).
6. A. Mine, N. Hino, and M. Ueda, Studies on the herbicidal properties of bentazon under paddyfield conditions, I. Environmental and cultural factors influencing herbicidal activity, *Zasso-Kenkyu [Weed Res. (Japan)]*, **17**, 64 (1973).
7. A. Mine, N. Hino, M. Ueda, and S. Matsunaka, Studies on the herbicidal properties of bentazon under paddyfield conditions, II. Absorption and translocation in plants and behaviors in soil, *Zasso-Kenkyu [Weed Res. (Japan)]*, **18**, 5 (1974).
8. A. Mine and S. Matsunaka, Mode of action of bentazon. Effect on photosynthesis, *Pestic. Biochem. Physiol.* **5**, 444–450 (1975).
9. A. Mine and S. Matsunaka, Mode of action and selective mechanism of bentazon, *Proc. Weed Soc. Japan, 12th Meet.*, 82 (1973).
10. W. Roth, Étude compareé de la réaction du mäis et du Blé a la Simazine, substance herbicide, *Compt. Rend.* H. **245**, 942 (1957).
11. R. H. Hamilton, Tolerance of several grass species to 2-chloro-*s*-triazine herbicides in relation to degradation and content of benzoxazinone derivatives, *J. Agr. Food Chem.* **12**, 14 (1964).
12. R. H. Shimabukuro, H. R. Swanson, and W. C. Walsh, Glutathione conjugation, atrazine detoxication mechanism in corn, *Plant Physiol.* **46**, 103 (1970).
13. R. Y. Yih, D. H. McRae, and H. F. Wilson, Mechanism of selective action of 3′,4′-dichloropropionanilide, *Plant Physiol.* **43**, 1291 (1968).
14. S. Matsunaka, Activation and inactivation of herbicides by higher plants, *Res. Rev.* **25**, 45 (1969).
15. E. W. Stoller and L. M. Wax, Amiben metabolism and selectivity, *Weed Sci.* **16**, 283 (1968).
16. G. R. Stephenson and S. K. Ries, The movement and metabolism of pyrazon in tolerant and susceptible species, *Weed Res.* **7**, 51 (1967).
17. S. J. Zaunbrecher and R. L. Rogers, Absorption and translocation of ^{14}C-bentazon (BAS-351-H) by soybean and cocklebur, *Proc. S. Weed Sci. Soc. Amer., 26 Meet.* 379 (1973).
18. R. M. Hayes and L. M. Wax, Absorption, translocation and metabolism of bentazon in soybean cultivars, *Abstr. Meet. Weed Sci. Soc. Amer., 1973 Meet.* 60 (1973).
19. S. Otto and N. Dresher, Trials on ^{14}C-labeled bentazon (BAS-3510H) in soya beans, *Laboratory report No. 1013, BASF Agric. Research Station*, (1971).
20. J. Kimura and H. Chiba, *J. Sci. Soil and Manure (Japan)* **17**, 479 (1943).
21. S. Goodwin, J. N. Shoolery, and L. F. Johnson, Nuclear magnetic resonance spectra of alkaloids. I. The complete structure of lunacrine and lunine, *J. Amer. Chem. Soc.* **81**, 3065 (1959).
22. J. R. Merchant and D. S. Chothia, Reactions of nitriles. Part VIII. Synthesis of 2,3-dihydroquinolin-4(1H)-ones, *J. Chem. Soc. Perkin I.* 932 (1972).