

Absorption, Translocation, and Residue of ¹⁴C-ZJ0273 in Oilseed Rape

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A study was conducted to investigate the absorption and translocation of a novel herbicide ¹⁴C-ZJ0273 in the seedlings and the residue in the mature plants of oilseed rape. Total absorption of ¹⁴C-ZJ0273 into seedlings and mature plants of oilseed rape reached 39.7% and 34.2% at the end of the experiments, respectively. Movement of the absorbed ¹⁴C in the plants was both acropetal and basipetal. However, more than 89.2% of the absorbed ¹⁴C remained in the treated leaf of the tested plants. The distribution of the absorbed ¹⁴C in the seedlings followed the order treated leaf (TL) > leaves above the treated leaf (LATL) > leaves below the treated leaf (LBTL) > roots and stalks, while in the mature oilseed rape, the order followed was TL > LATL > LBTL and stems > seed capsules, seed, and roots > branches. Only the parent compound ZJ0273 was found in the extractable residue of the oil rape seeds with a concentration of 0.09 mg per kg dry weight. The residue level in oil seeds suggested a low dietary exposure for humans if recommended application rates were followed.

KEYWORDS: Absorption; translocation; residue; herbicide; ZJ0273

INTRODUCTION

Oilseed rape (Brassica napus L.) is an important source of edible oil in the world, and rape oil accounts for about 35% of the edible oil in China (1). However, weed infestation in oilseed rape fields could cause yield reduction from 15 to over 50% (2). Consequently, different herbicides such as benazolin, trifluralin, metazachlor, propyzamide, and carbetamide, among others, have been used to control weeds in rape fields during the pre- and/or early postemergence periods. A novel herbicide, ZJ0273 (propyl 4-(2-(4,6-dimethoxypyrimidin-2-yloxy) benzylamino)benzoate), is being developed for weed control primarily in oilseed rape production (3). At an application rate of 60-150 g ai h⁻¹, ZJ0273 was found to effectively control many monocotyledonous and dicotyledonous weeds, including crickweed (Malachium aquaticum L.), chickweed (Stellaria media L.), redroot amaranth (Amaranthus retroflexus L.), equal alopecurus (Alopecurus aequalis Sobol.), japanese alopecurus (Alopecurus japonicus Steud.), annual bluegrass (Poa annua L.), common polypogon (Polypogon fugax Nees ex Steud.), keng stiffgrass (Sclerochloa kengiana (Ohwi) Tzvel.), and spinefruit buttercup (Ranunculus muricatus L.), with an efficacy of over 80% (4, 5).

Herbicides, especially novel herbicides, must be evaluated before wide use to ensure that they have no harm to human health or the environment. In addition, as oilseed rape is an important source of edible oil for human consumption, safety assessment must consider pesticide residue in the oil rape seeds, and an understanding of the absorption, distribution of residues, and the nature of the terminal residue (6). The environmental behavior of ZJ0273 in soils was addressed in recently published studies (7, 8). However, the absorption, translocation, distribution, and residue of ZJ0273 in the oilseed rape still remain unknown. The objectives of this study were to evaluate the foliar absorption and translocation of ¹⁴C-ZJ0273 in oilseed rape seedlings, the distribution of herbicide residue in the different parts of mature oilseed rape plants, and the terminal residue of ZJ0273 in the oil rape seeds under simulated field conditions.

MATERIALS AND METHODS

Chemicals. The synthesis of [A ring-U-¹⁴C] ZJ0273 (propyl 4-(2-(4,6-dimethoxypyrimidin-2-yloxy)benzylamino)[*phenyl*-U-¹⁴C]benzoate, **Figure 1**) was reported previously (9). The specific activity was 4.8×10^4 Bq mg⁻¹. The radiochemical purity and chemical purity were >98%. The cocktail ingredients 2,5-diphenyloxazole (PPO) and 1,4-di-[2'-(5'-phenyloxazoly]]-benzene (POPOP) were both of scintillation grade. Dimethylbenzene, dichloromethane, acetone, glycol-ether, and ethanol-amine were of analytical grade. Methanol, ethyl acetate, and cyclohexane used in the chromatograph analysis were of chromatographic grade.

Seedling Absorption and Translocation Experiment. Seeds of oilseed rape (HO 605) were germinated at 25 °C in Petri dishes containing filter papers moistened with distilled water. After germination, seeds were sowed in 500-mL pots containing a fluvio-marine yellow loamy soil (pH 7.02, organic matter content of 30.5 g kg⁻¹, and sand content of 20.8%). After emergence, plants were thinned to one seedling per pot and cultivated under laboratory conditions (25–28/22–25 °C, day/night; humidity, 80%; light, 12 h/12 h). Plant containers were watered manually as needed until treatment.

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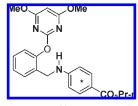


Figure 1. Structure of [A ring-U-¹⁴C]ZJ0273 (propyl 4-(2-(4,6-dimethoxypyrimidin-2-yloxy)benzylamino)[*phenyl*-U-¹⁴C]benzoate).

The methods of treatment and analysis were modified according to the procedure of Lycan et al. (10). The seedlings of the oilseed rape at the three-leaf stage were selected for treatment. The second fully expanded leaf of the selected plants was treated with 20 µL of spotting solution containing 0.25% nonionic surfactant Tween-80 and a total radioactivity of 4445 Bq of [A ring-U-14C]ZJ0273. Prior to treatment, the soil surface of the pots were covered with filter papers to prevent the spotting solution from contaminating the soil. After treatment, the seedlings were cultivated under normal conditions until sampling. Seedlings of oilseed rape were harvested in batches of 10 plants at 3, 9, 12, 24, 48, 72, and 96 h after treatment (HAT). The treated leaves were excised off the plants and rinsed with acetone for 30 s at a flow rate of 5 mL min⁻¹ using a Waters 600E pump (Waters Co., Massachusetts, USA) to remove the nonabsorbed herbicide. The leaf washes were directly collected in 20-mL glass scintillation vials. After acetone had evaporated in the fume hood, 10 mL of scintillation cocktail A (5 g of PPO + 0.5 g of POPOP + 650 mL of dimethylbenzene + 350 mL of glycol-ether) was added. The ¹⁴C radioactivity (dpm) was measured using a liquid scintillation counter (LSC: Wallac 1414, Turku, Finland). In a preliminary experiment, leaves were spotted with ¹⁴C-ZJ0273 and sampled after 10 s to evaluate the efficiency of the leaf wash technique (11). The method removed about $98.5 \pm 2.5\%$ of nonabsorbed ¹⁴C-ZJ0273. The treated plants were divided into five parts: treated leaf (TL), leaves above the treated leaf (LATL), leaves below the treated leaf (LBTL), stalks, and roots. After drying to a constant weight in a drying oven at 80 °C, all of the plant parts were combusted using a biological oxidizer (OX-600 Biological Oxidizer, R. J. Harvey Instrument Co., New Jersey, USA). The time duration of combustion was 4 min, with the combustion and catalyst zone temperatures at 900 and 680 °C, respectively. The released ¹⁴CO₂ from sample combustion was trapped in 15 mL of scintillation cocktail B (5 g of PPO + 0.5 g of POPOP + 600 mL of dimethylbenzene + 225 mL of glycol-ether + 175 mL of ethanolamine). The radioactivity was determined by LSC to quantify the absorbed ¹⁴C-ZJ0273 in plant tissues. The recovery efficiency of the above combustion procedure, as determined by combusting a known amount of ¹⁴C-ZJ0273 standard, was >90%. Foliar absorption was calculated by dividing the amount of ¹⁴C recovered from the analyzed plant part by the applied ¹⁴C activity. Translocation of ¹⁴C was expressed as the percentage of activity in a given tissue over the total absorbed radioactivity.

Mature Plant Experiment. Eighty 3-leaf oilseed rape seedlings were transplanted to plastic pots (35 cm in diameter and 30 cm in height) containing the fluvio-marine yellow loamy soil and cultivated under open outdoor conditions similar to field conditions, covering only when necessary to prevent rain or snow. A single plant was planted in each container. Oilseed rape seedlings in 60 containers were individually treated with 20 μ L of spotting solution containing 34500 Bq of [A ring-U-¹⁴C] ZJ0273, and the application rate (0.72 mg per plant) was consistent with the recommended field use rate (60-150 g ai h⁻¹, assuming 11 plants per m²). In order to maximize the detection of radiolabeled herbicide and/ or its metabolites, we chose not to spray plants with the nonradiolabeled herbicide to avoid dilution effects. Prior to treatment, the soil surface of the pots were covered with filter papers to prevent the spiking solution from contaminating the soil. The remaining 20 pots were used as the control. All of the plants were harvested at the mature stage (238 days after treatment). The individual oilseed rape plant was divided into the following parts: treated leaf, leaves above treated leaf, leaves below treated leaf, stems, branches, seed capsule, seeds, and roots. Seeds of the individual rape plant were ground to a homogenized powder under liquid nitrogen using a mortar and pestle. Other plant parts were dried to a constant weight at 80 °C in a drying oven, weighed, and finally ground to a homogenized powder in an Udy cyclone mill (FZ102, Qi-jia-wu instrument Co., Hebei, China). The different plant parts from the control were weighed after drying. Three 200-mg aliquots (oven-dry weight) were removed from each plant part and were combusted as decribed above. ¹⁴CO₂ from the combustion was trapped in 15 mL of scintillation cocktail B, and the radioactivity was determined on a Quantulus 1200 ultralow liquid scintillation spectrometer (ULLSS; Wallac, Turku, Finland) to assess the absorbed ¹⁴C-ZJ0273. Allocation of ¹⁴C was expressed as the percentage of radioactivity in a given plant part to the total absorbed radioactivity. ¹⁴C tissue concentration was expressed as radioactivity per gram dry weight (Bq g⁻¹).

Analysis of Residue in Oil Rape Seeds. To evaluate the residue of ¹⁴C-ZJ0273, the remaining seed powder from 20 random plants were combined and transferred to 100-mL capped centrifuge tubes with 10 g of powder per tube. The powder was extracted by shaking with 50 mL of 80% methanol for 2 h once, followed by 50 mL of pure methanol for 2 h twice, and then by 50 mL of chloroform for 2 h once. The extracts from each step were filtered through a three-layered gauze, centrifuged at 4000 rpm for 30 min, and the supernatant was collected. The 80% methanol extract was evaporated to remove all of the methanol on a vacuumed rotary evaporator at 40 °C (R-202, Shanghai Shensheng Bio-Tech, Shanghai, China). The aqueous phase was adjusted to pH 3.0 with diluted HCl, partitioned three times with an equal volume of dichloromethane, and the aqueous layer (below background) was discarded. All of the organic phases from the extracts were combined and evaporated to near dryness under vacuum on a rotary evaporator. The residue was dissolved in 25 mL of ethyl acetate/cyclohexane (1:1) and further condensed to 5 mL. After centrifugation at 16000 rpm for 30 min, the samples were purified by gel permeation chromatography (GPC) equipped with a 30×400 mm column packed with bio beads S-X3 (200-400 mesh, Bio-Rad, California, USA). The column was first eluted with ethyl acetate/cyclohexane (1:1) at a flow rate of 2.0 mL min⁻¹ controlled using a Waters 600E pump (Waters Co., Massachusetts USA). The above sample $(2.5 \text{ mL} \times 2)$ was loaded onto the GPC column, and the eluted fractions were collected 10 mL per tube, and 1.0 mL aliquots were taken to detect the radioactivity on a ULLSS. All of the radioactive fractions were combined and evaporated to near dryness under vacuum at 40 °C on a rotary evaporator. The residue was dissolved and brought to 1.5 mL with methanol. A subsample was centrifuged at 18000 rpm for 30 min, and filtered through a 0.22- μ m pore size filter (Millipore Co., Massachusetts, USA). An aliquot of 40 μ L sample was injected into an HPLC system equipped with a reversed-phase Diamonsil C18 column (5- μ m, 4.6 \times 250 mm, Dikma Technologies), a Waters 600 multisolvent delivery unit, and a Waters 996 photodiode array detector operating at 301 and 254 nm. With the column temperature maintained at 30 °C, the column was eluted with mobile phases A (double distilled water with 0.1% acetic acid) and B (methanol with 0.1% acetic acid) using a gradient mode (%A/min: 80/0, 25/40, 25/80, 0/85, 0/ 90, 100/105, and 80/110) at a flow rate of 1 mL min⁻¹. The eluted fractions were collected in 20 mL scintillation vials (1.0 mL per vial), and after the addition of 10 mL of scintillation cocktail A to each vial, the radioactivity was quantified on a ULLSS to determine the radioactive peak. The parent compound of ZJ0273 was identified by the retention time using a technical standard. The standard unlabeled ZJ0273 was added to a subsample of the extract, and ${\rm ^{14}C}$ and absorbance at 301 nm were compared in the same chromatogram. During the process of analysis, the radioactivity of the discarded fractions was detected to ensure that only the fractions with the radioactivity lower than background were discarded.

The amount of 14 C radioactivity in the remaining solid residue after the four-step extraction was determined by sample oxidation and referred to as initially bound throughout this article. Three 1-g aliquots of the solid residue were combusted as described above, and the released 14 CO₂ was trapped in 15 mL of scintillation cocktail B. The radioactivity was determined on the ULLS. The above residue analysis was repeated three times.

Statistical Analysis. The radioactive data had deducted the background (20 dpm for ULLSS and 60 dpm for LSC). All data were analyzed using SigmaPlot 9.0 (Systat Sofeware, Richmond, CA, USA). Absorption, translocation, and residue analysis data were subjected to an analysis of variance (ANOVA). LSD test was used to test statistical significance (p < 0.05).

RESULTS AND DISCUSSION

Foliar Absorption and Translocation of ¹⁴**C-ZJ0273 in Seedlings.** The ¹⁴C radioactivity recovered in TL, LATL, LBTL, and roots were summed to derive the total absorption for a single plant. At 3 HAT, 5.83% of the applied ¹⁴C-ZJ0273 was absorbed into the seedlings of oilseed rape. Foliar absorption of ¹⁴C-ZJ0273 into the seedlings increased with the sampling time, reaching 39.7% at 96 HAT (Figure 2).

More than 89.2% of the absorbed ¹⁴C remained in the treated leaf of the oilseed rape seedlings (Table 1). Therefore, only an extremely relatively small portion of the absorbed ¹⁴C moved out of the treated leaf to other parts of the plant, which indicated that the mobility of ZJ0273 and/or its metabolites within young oilseed rape plants was rather limited. In addition, a moderately greater fraction of the absorbed ¹⁴C was found in the treated leaf at 96 HAT (95.7%) than that at 6 HAT (89.2%) (Table 1), the cause of which may lie in the increase in ¹⁴C absorption that may have changed the proportion of ¹⁴C in the treated leaf in relation to the other fractions. Other researchers have reported a similar observation that the majority of foliar absorbed ¹⁴C-sulfosulfuron, or -bispyribac-sodium remained in the treated leaf of various plant species even after 72 h or longer. For example, Olson et al. reported that 90 to 97% of absorbed ¹⁴C-sulfosulfuron remained in the treated leaves of wheat (Triticum aestivum L.) and other grass species after 96 h (12).

Translocation of ¹⁴C was acropetal and basipetal in oilseed rape seedlings. At 96 HAT, a lower amount of the absorbed ¹⁴C was found in LATL (2.19%), LBTL (1.56%), stalks (0.32%), and roots (0.29%) of oilseed rape seedlings compared to 6.80%, 3.09%, 0.80%, and 0.87% at 3 HAT, respectively, due to less increase in the translocation rate than in total absorption rate (**Table 1**). At 96 HAT, the allocation of ¹⁴C in LATL and LBTL was far greater than that in the stalks and roots of oilseed rape (**Table 1**), which indicated that ZJ0273 and/or its metabolites was more readily accumulated in the leaves. No significant difference was found between the stalks and the roots at 96 HAT.

¹⁴C Allocation in Mature Plants of Oilseed Rape. According to a LSD test (p < 0.05), no significant differences were observed in the total dry weight of the plants and the yield of seeds between the treated oilseed rape and the control (data not shown), which indicated that the herbicide had no inhibition effect on the oilseed rape in our experiment. The results of a field study carried out by Fu et al. also showed that rape cultivars from different areas were not injured and that the yield improved compared to the control when ZJ0273 10% SC was applied (*13*).

The total absorption of ZJ0273 into the mature oilseed rape reached $34.2 \pm 2.5\%$ at harvest time (**Table 2**). That was slightly lower than the absorption at 96 HAT into the seedlings ($39.7 \pm$ 1.71% of applied). The different absorption might result from the different growth conditions such as temperature, relative humidity, soil moisture level, and irradiance level among other factors between the laboratory and the field.

About 94.0% of the absorbed ¹⁴C remained in the treated leaf of the mature oilseed rape, which was lower than that in the oil rape seedlings at 96 HAT. Compared to the ¹⁴C allocation in the seedlings at 96 HAT, a greater amount of ¹⁴C was present in the roots (0.46%) of the mature oilseed rape at harvest, while a smaller fraction was associated with LBTL (1.31%). The proportion of the absorbed ¹⁴C in LATL (1.79%) was greater than the other parts except for the treated leaf, and the branches (0.34%) accumulated the smallest percentage of the absorbed ¹⁴C. No significant difference was found in the allocation of ¹⁴C between LBTL (1.31%) and stems (1.29%), or among seed capsules (0.50%), seeds (0.51%), and the roots (0.46%) (**Table 2**). Overall,

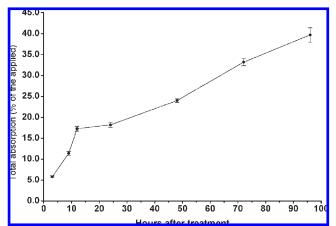


Figure 2. Total absorption of 14 C-ZJ0273 in seedlings of oilseed rape. Each point is the mean of 10 replications \pm standard error of the mean.

the concentration of ¹⁴C in the different parts followed the order LBTL > LATL > stems > roots, branches > seeds and seed capsule, significantly (**Table 2**). Thus, conclusions could be drawn that ¹⁴C-ZJ0273 and/or its metabolites were more readily accumulated in the leaves than in the other parts of an oilseed rape plant.

Residue in Seeds. The HPLC column separated ZJ0273 and/or its probable metabolites (Figure 3). The radioactive chromatogram was plotted on the basis of the retention time and radioactivity of all eluted fractions (Figure 4). Only detection of radioactive $> 2 \times$ the background (20 dpm for ULLSS) was considered a radioactive peak. Thus, only one radioactive peak was found in the radioactive chromatogram of the HPLC eluate (Figure 4), and the retention time of the peak coincided with that of the authentic standard ZJ0273, suggesting that the primary radioactive component in oilseed rape seeds was [A ring-U-¹⁴C] ZJ0273. Fortification of a subsample of the same extract with nonlabeled ZJ0273 was carried out to compare radioactivity distribution and UV absorbance at 301 nm under the same conditions, and the results further confirmed the above assignments of the radioactivity peak to the parent compound. Further analysis of LC/MS showed that the primary radioactive peak in the extracted residue was ZJ0273, and the identification was based on characteristic fragment ion (m/z 245) and two adduct ions (m/z 424, M + H and m/z 446, M + Na) (data not shown). Radioactive quantification revealed that about $87.5 \pm 2.5\%$ of the radioactivity in the seeds was found in the extracts and that essentially all of the extractable residue was the parent compound ZJ0273. The radioactivity remaining in the solid residue was about $8.5 \pm 0.7\%$ of the total radioactivity in the seeds. The nature of the unextractable residue was not further characterized in the current study because of the low level of the initially bound residue. The concentration of the parent ZJ0273 in the oil rape seeds was 0.09 mg per kg dry weight. The residue level of ZJ0273 in oil rape seeds under field conditions was less than 0.006 mg kg¹⁻, after a single spray application of 10% ZJ0273 EC at the dosage of 600 and 900 mL hm⁻² after the transplant of oilseed rape seedlings in the field (5). The reported level was lower than those found in the current study. The differences may be attributed to the use of the multisolvent, multistep extraction procedure in this study and the improved sensitivity due to the use of the ¹⁴C-labeling technique. Generally, the herbicide and its metabolites are transported out of the treated leaf to the seeds usually through the phloem or/and xylem, and the most important chemical properties in determining the transport pattern in plants are pK_a and Kow values (14). Various groups of herbicides

| | hours after treatment | | | | | | | | | | |
|-----------------|---|--|---|---|---|---|---|--|--|--|--|
| | 3 | 9 | 12 | 24 | 48 | 72 | 96 | | | | |
| TL | 89.2 ± 0.6 | 93.4 ± 0.1 | 94.1 ± 0.1a | $94.1\pm0.2a$ | $94.4\pm0.1a$ | $95.5\pm0.2a1$ | $95.7\pm0.2a1$ | | | | |
| LATL | 6.80 ± 0.33 | $4.07\pm0.11\text{b}$ | $3.69\pm0.11b$ | $3.56\pm0.08\mathrm{b}$ | 3.21 ± 0.15 | $2.31\pm0.09\text{b1}$ | $2.19\pm0.06\text{b1}$ | | | | |
| LBTL | 2.28 ± 0.27 | 1.85 ± 0.09 | $1.62\pm0.04c$ | $1.65\pm0.06\mathrm{c}$ | $1.62\pm0.05\mathrm{c}$ | $1.61\pm0.07c$ | $1.56\pm0.06c$ | | | | |
| stalks roots | $\begin{array}{c} 0.80 \pm 0.05 \\ 0.87 \pm 0.06 \end{array}$ | $\begin{array}{c} \textbf{0.29} \pm \textbf{0.03d} \\ \textbf{0.48} \pm \textbf{0.05} \end{array}$ | $\begin{array}{c} \textbf{0.27} \pm \textbf{0.02d} \\ \textbf{0.34} \pm \textbf{0.01e} \end{array}$ | $\begin{array}{c} \textbf{0.29} \pm \textbf{0.03d} \\ \textbf{0.37} \pm \textbf{0.02e} \end{array}$ | $\begin{array}{c} {\rm 0.22 \pm 0.03d} \\ {\rm 0.36 \pm 0.02e} \end{array}$ | $\begin{array}{c} \textbf{0.25} \pm \textbf{0.01d} \\ \textbf{0.35} \pm \textbf{0.01e} \end{array}$ | $\begin{array}{c} \textbf{0.32} \pm \textbf{0.04d} \\ \textbf{0.29} \pm \textbf{0.02d} \end{array}$ | | | | |

^a Data are the mean of 10 replications \pm standard error of the mean. Means of plant parts for each day followed by the same letter are not significantly different according to ANOVA (P < 0.05).

Table 2. Absorption, Distribution, and Concentration of ZJ0273 in the Mature Plants of Oilseed Rape^a

| | TL | LATL | LBTL | stems | branches | seed capsule | seeds | roots | |
|--|-----------------------|----------------|----------------|----------------|--------------------------|-----------------------|-----------------|----------------|--|
| absorption (% of the absorbed ¹⁴ C) | 34.2 ± 2.5 | | | | | | | | |
| allocation (% of the absorbed ¹⁴ C) | 94.0 ± 0.5 | 1.79 ± 0.03 | $1.31\pm0.04a$ | $1.29\pm0.06a$ | 0.34 ± 0.01 | $0.50\pm0.01\text{b}$ | $0.51\pm0.02b$ | $0.46\pm0.01b$ | |
| concentration (Bq g^{-1}) | 13879.76 ± 559.31 | 16.38 ± 1.42 | 32.25 ± 2.47 | 8.50 ± 0.31 | $6.61\pm0.29 \mathrm{c}$ | $5.49\pm0.13d$ | 5.22 ± 0.14 d | $6.81\pm0.36c$ | |

^a Data are the mean of 60 replications \pm standard error of the mean. Means of plant parts followed by the same letter are not significantly different according to ANOVA (P < 0.05).

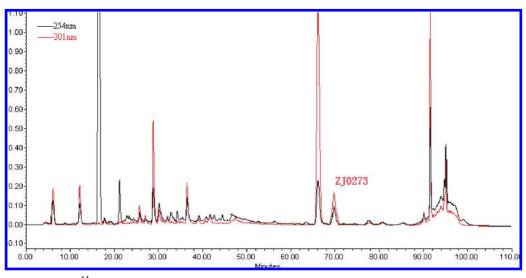


Figure 3. HPLC chromatogram of ¹⁴C-ZJ0273 in oil rape seeds.

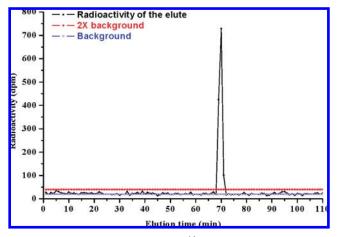


Figure 4. Radioactive chromatogram of ¹⁴C-ZJ0273 in oil rape seeds.

have been superimposed on a model proposed by Bromilow et al., and the results showed that a compound with log Kow >4 may be considered nonmobile in phloem and xylem (15). As ZJ0273 is relatively lipophilic (log P = 4.1), it was not surprising that the mobility of ZJ0273 in phloem and xylem was weak, which accounts for the low ZJ0273 residue level in oil rape seeds. As a herbicide used mainly at the seedling stage of oilseed rape, the residue of ZJ0273 in the oil rape seeds was most likely derived from the translocation of ZJ0273 and/or its metabolites. Therefore, weak mobility of the radioactivity from [A ring-U-¹⁴C] ZJ0273 may be an indication of low residue levels derived from ZJ0273 under field conditions.

As the residue of [A ring-U-¹⁴C]ZJ0273 in the seeds existed predominantly as the parent compound, safety analysis should focus on ZJ0273. Preliminary studies on the mode of action of ZJ0273 showed that the herbicide inhibited the synthesis of the branched-chain amino acids (leucine, isoleucine, and valine) within a susceptible plant in vivo and that the inhibition effect was counteracted by the addition of branched-chain amino acids, which indicated that ZJ0273 was the inhibitor of ALS-acetolactate synthase (16). As ALS is present in plants as well as in bacteria, fungi, algae, and yeasts but not in animals, the ALS-inhibiting herbicides are generally thought to have low toxicity to mammals (17). The two-year maximal no-effect dosages of ZJ0273 on Sprague-Dawley rats were 145.6 and 193.4 mg kg⁻¹ d⁻¹ for males and females, respectively, and no detectable carcinogenic effect was found. The variety HO 605, used in our experiment, is a high-oil variety of winter oilseed rape

with an initial oil content of 49.78% and a seed oil of 40.93% in processing (18, 19). According to the recommended daily consumption amount of cooking oil of 25–30 g per person (20), the daily consumption could equal to 61–73 g d⁻¹ in rape seeds, provided that only the rapeseed oil is consumed. Thus, the maximum residue of ZJ0273 in the daily edible oil would be in the range of 5.5–6.6 μ g per person. That is much smaller than the two-year maximal no-effect dosages of ZJ0273 for rats. Therefore, results from this study suggest that the residue level of ZJ0273 in rape seeds would not reach a harmful level when ZJ0273 is applied at the recommended dosage.

In summary, the total absorption of the applied ¹⁴C-ZJ0273 into the seedlings of oilseed rape reached 39.7% at 96 HAT and 34.2% in mature oil rape at harvest time. In both experiments, more than 89.2% of the absorbed ¹⁴C remained in the treated leaf, and the leaves accumulated more ¹⁴C than the other parts of the plants. At the rate used, the herbicide had no inhibition effect on the dry weight of the oilseed rape plants and seeds. The residue of ZJ0273 in the seeds existed primarily in the parent form, which accounted for 87.5 \pm 2.5% of the total ¹⁴C radioactivity in the seeds. Integrative analysis of potential exposure to ZJ0273 via the consumption of rape seed oil indicated that ZJ0273 used on oil rape plants at the recommended rates should not constitute a potential hazard to humans.

ABBREVIATION USED

TL, treated leaf; LATL, leaves above the treated leaf; LBTL, leaves below the treated leaf; HPLC, high-performance liquid chromatography; LSC, liquid scintillation counter.

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