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CHANGES IN SOIL BIOLOGICAL PROPERTIES WITH THE ADDITION OF METSULFURON-METHYL HERBICIDE*

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Abstract: An incubation study was conducted to investigate the effects of metsulfuron-methyl herbicide on the soil microbial biomass in loamy sand soil. The herbicide was applied to the soil at four concentrations: control, 0.01, 0.10, and 1.00 $\mu g \cdot g^{-1}$ soil. Determinations of microbial biomass-C and microbial biomass-N contents were carried out 1, 3, 5, 7, 10, 15, 25, and 45 days after herbicide application. In comparison to untreated soil, the microbial biomass-C and biomass-N decreased significantly in soils treated with herbicide at concentrations of 0.1 and 1.0 $\mu g \cdot g^{-1}$ soil within the first 7 days of incubation. The application of metsulfuron-methyl herbicide to the soil reduced the C_{mic}/C_{org} and N_{mic}/N_{total} percentages, which decreased with increasing application rate of metsulfuron-methyl herbicide. Compared to the untreated control, a marked increase in the microbial biomass C: N ratio was observed in the herbicide treated soil. This effect was transitory and was significant only at the higher rates of metsulfuron-methyl.

Key words: metsulfuron-methyl, microbial biomass C, microbial biomass N, C_{mic}/C_{org} ratio, N_{mic}/N_{total} ratio, C_{mic}/N_{mic} ratio

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INTRODUCTION

The maintenance of soil fertility depends on the size and activity of soil microbial biomass (Alexander, 1977), which is of fundamental importance in the biological cycles of almost all major plant nutrients (Robert and Chenu, 1992). Although the soil microbial biomass represents only a small fraction of the total amount of soil C, N, P and S, it has a relatively rapid turnover (Amato and Ladd, 1980; Marumoto et al., 1982). The biomass has a multiple role in soil, affecting the decomposition and turnover of organic matter, nutrient immobilization and cycling, root physiology and soil structure (Vaughan and Malcolm, 1985).

Herbicides affect various soil microbial processes (Tyler, 1980), inhibit decomposition (Grossbard and Wingfield, 1978) and depending on type and rate of application, can alter the biomass quantitatively and qualitatively in both the short and long-term (Anderson et al., 1981). Much work concerning the toxicity of sulfonylurea herbicides on weeds and crops has been reported (Bayer, et al., 1987; John et al., 1993), but there are few reports about their direct effects on soil microbial biomass and activities.

The sulfonylurea herbicides are a relatively new group of compounds, which control broad-leaved weeds and some grasses in cereal crops (Blair and Martin, 1988), at very low application rates ($2-75$ $g \cdot ha^{-1}$) (Brown, 1990). Metsulfuron-methyl is sulfonylurea herbicide, with high herbicidal activity and very low mammalian toxicity and is widely used in agriculture (James, 1990). The half-life of metsulfuron-methyl at different soil water content and temperatures is 8 to 36 days (James et al., 1995). The use of pesticides is thought to be harmful to

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microorganisms and their activities contributing to soil fertility and is therefore avoided (Wolf, 1977). Ismail et al., (1996) observed that microbial biomass in clay loam soil increased with herbicide (metsulfuron-methyl) treatment during the first 9 days of incubation, but declined from day 19 onward. However, in sandy loam soil, the biomass decreased with an increase of herbicide concentration on day 1, but increased thereafter. Perucci and Scarponi (1996) showed that rimsulfuron herbicide decreased the microbial biomass carbon at 10 FR and 100 FR within the first 10 days.

Thus anything, like herbicide, that disrupts microbial activity in soils, could be expected to affect the long-term soil productivity and would have serious consequences. The present investigation aimed at assessing the influence of metsulfuron-methyl on the size of soil microbial biomass-C (C_{mic}), microbial biomass-N (N_{mic}), microbial biomass C/organic C (C_{mic}/C_{org}) percent, microbial biomass N/total N (N_{mic}/N_{total}) percent and the ratio of C_{mic}/N_{mic} in loamy sand soil.

MATERIALS AND METHODS

A laboratory incubation experiment was conducted using loamy sand soil with 17.6 $g \cdot kg^{-1}$ total organic carbon, 1.58 $g \cdot kg^{-1}$ total nitrogen, 22.4% soil moisture at − 33 kPa, and pH of 6.27. The soil was collected from the surface layer (0 − 20 cm) from Hangzhou, Zhejiang Province, China.

After sampling and preparation, the soil sample was divided into four sub-samples. One sub-sample was used as control, and the others were treated with metsulfuron-methyl herbicide as solutions of metsulfuron-methyl at concentrations of 0.5, 5.0, and 50 $\mu g \cdot ml^{-1}$.

The herbicide was incorporated into the soil sub-sample by adding 48 ml of the methanolic solution of metsulfuron-methyl to 120 g of air-dried soil sub-sample. An equal volume of methanol instead of the herbicide was added to the control soil.

After the complete removal of the methanol by evaporation at room temperature, each of the 120 g soil samples was divided into 24 portions each with 5 g soil for transfer into beakers containing 95 g fresh soil (oven dried) and homogenized. With this procedure, sub-samples for application rates of to 0.01(L_1), 0.10 (L_2), and 1.00 (L3)$\mu g \cdot g^{-1}$ soil were obtained.

Soil moisture was adjusted to 60% water content at − 33 kPa and incubated in the dark at 25 ± 1℃. The beakers were removed from the incubator every day and brought to the original weight by adding the required amount of distilled water. Three beakers each for control and treated soils were removed and analyzed for C_{mic} and N_{mic} 1, 3, 5, 7, 10, 15, 25 and 45 days after metsulfuron-methyl treatment.

Soil samples for determination of microbial biomass C were extracted by a fumigation-extraction (FE) method (Vance et al., 1987) and the organic carbon in the soil extracts was measured using an automated total organic carbon analyzer (Wu et al., 1990). Soil samples for determination of microbial biomass N were extracted by a fumigation-extraction (FE) method (Brookes et al., 1985b) and the total nitrogen in the soil extracts was measured after Kjeldahl digestion (Brookes et al, 1985a).

Water contents at applied pressure of − 33 kPa were determined using a pressure membrane system similar to that described by Heining (1963). The pH (in water, 1:2.5) of the soils was measured with a pH meter. Total N was determined by Kjeldahl method and total organic carbon by Walkley-Black procedure (Jackson, 1958).

Data were examined by analysis of variance completely randomized and Duncan's multiple range tests using statistix software (CoStat software 1990).

RESULTS

1. The effect of metsulfuron-methyl on soil microbial biomass-C (C_{mic})

Fig. 1 on the responses of the soil microbial biomass-C content to the herbicide treatments shows that the C_{mic} contents were not affected significantly at 0.01$\mu g \cdot g^{-1}$ soil application rate. But when metsulfuron-methyl was applied at the rate of 0.1and 1.0 $\mu g \cdot g^{-1}$ soil, the reduction in the size of C_{mic} became significant, especially within the first 7days, as compared with the control:

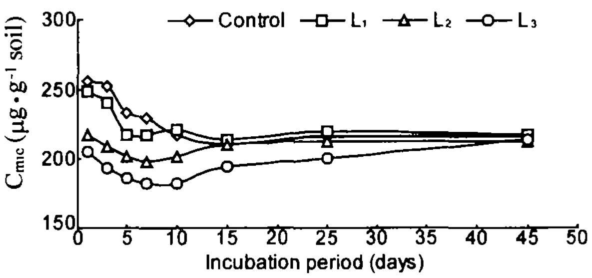

Fig. 1 The effect of metsulfuron-methyl on soil microbial biomass-C (C_{mic})
L_1(0.01 μg·g^{-1}), L_2(0.1 μg·g^{-1}), and L_3(1.0 μg·g^{-1})

The results revealed that when metsulfuron-methyl was added at the rate of 0.01 μg·g^{-1} soil, it reduced the C_{mic} contents by 2.9, 4.5, 6.6, and 5.2% on the 1st, 3rd, 5th, and 7th day of incubation, respectively, as compared with the control. While a nominal increase of 1.9%, 1.8%, 1.9%, and 0.3% was noticed after 10, 15, 25, and 45 days of incubation, respectively, compared with the control. However, when it was applied at 0.1μg·g^{-1} rate, the decrease in C_{mic} was 15.2%, 16.8%, 13.3%, 13.6%, 7.1%, 0.1% (increase), 1.6%, and 1.7%, respectively, as compared with the control at the same incubation periods. The soil treated with 1.0 μg·g^{-1} soil with herbicide resulted in 19.7%, 23.1%, 20.1%, 20.4%, 15.9%, 7.3%, 7.1%, and 1.2% decline in C_{mic}, respectively, as compared with the control at the same incubation periods.

2. The effect of metsulfuron-methyl on soil microbial biomass-N (N_{mic})

Fig. 2 data on the effects of metsulfuron-methyl treatments on microbial biomass N is shows that the addition of metsulfuron-methyl at the rate of 0.01 μg·g^{-1}soil caused no significant change in N_{mic} contents. However, the 0.1 μg·g^{-1} soil application caused significant reduction in N_{mic} contents and its reduction with 1.0 μg·g^{-1} soil became highly significant, especially within first the 7 days of incubation, as compared with the control.

The results indicated that metsulfuron-methyl added at 0.01 μg·g^{-1} soil resulted in 6.1%, 10.8%, 8.8%, 6.4%, 2.7%, 2.9%, 2.7% and 2.6% reduction in N_{mic} after 1, 3, 5, 7, 10, 15, 25, and 45 days of incubation, respectively, as compared with the control. A marked decline in N_{mic} was also observed at 0.1μg·g^{-1} soil dosage, where it was reduced by 20.9%, 18.8%, 14.7%, 21.9%, 17.1%, 11.4%, 5.4%, and 2.8% at the same incubation periods, respectively, as compared with the control. However, 1.0 μg·g^{-1} soil herbicide decreased N_{mic} by 31.3%, 32.5%, 29.5%, 28.2%, 22.7%, 17.0%, 11.1%, and 10.9%, at the same incubation periods, respectively, compared to the control.

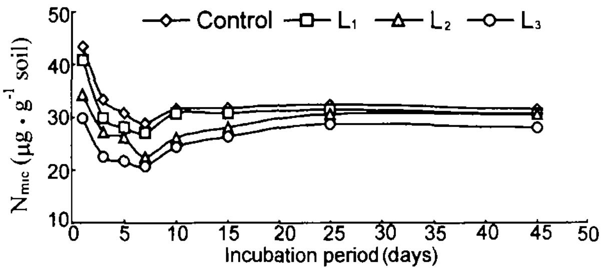

Fig. 2 The effect of metsulfuron-methyl on soil microbial biomass-N (N_{mic})
L_1(0.01 μg·g^{-1}), L_2(0.1 μg·g^{-1}), and L_3(1.0 μg·g^{-1})

3. Effect of metsulfuron-methyl on C_{mic}: C_{org} ratio

The application of metsulfuron-methyl herbicide to the soil reduced the C_{mic}/C_{org} percentage, which decreased with increasing application rate of metsulfuron-methyl herbicide (Fig. 3). Compared with the control, it was 4.8%, 3.8%, 9.3%, 9.8%, 0.51%, 0.46% (increase), 1.5% (increase), and 0.03% lower 1, 3, 5, 7, 10, 15, 25, and 45 days after incubation began, respectively; for the 0.01 μg·g^{-1} soil treatment, it was 16.3%, 19.1%, 14.5%, 18.2%, 10.7%, 6.6%, 6.9%, and 4.4% lower, respectively, with the 0.1 μg·g^{-1} soil treatment and 19.9%, 25.1%, 22.6%, 23.8%, 18.0%, 11.6%, 11.98%, and 0.97% lower, respectively, with the 1.0 μg·g^{-1} soil treatment.

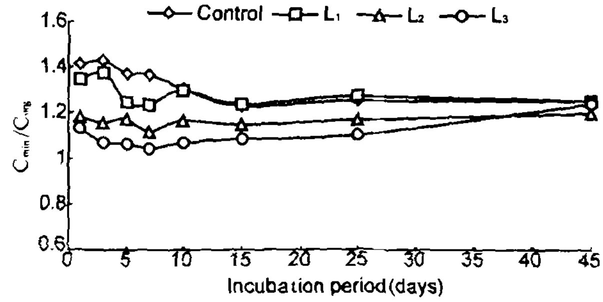

Fig.3 Effect of metsulfuron-methyl herbicide on microbial biomass C/ total organic C ratio (C_{mic}/C_{org} ratio) in a loamy sand soil

$L_1(0.01\ \mu g \cdot g^{-1})$; $L_2(0.1\ \mu g \cdot g^{-1})$; $L_3(1.0\ \mu g \cdot g^{-1})$

4. Effect of metsulfuron-methyl on N_{mic}: N_{total} ratio

The application of metsulfuron-methyl herbicide to the soil reduced the N_{mic}/N_{total} percentage (Fig. 4). There was consistent decrease in the N_{mic}/N_{total} percentage with increasing application levels of metsulfuron-methyl herbicide in soil.

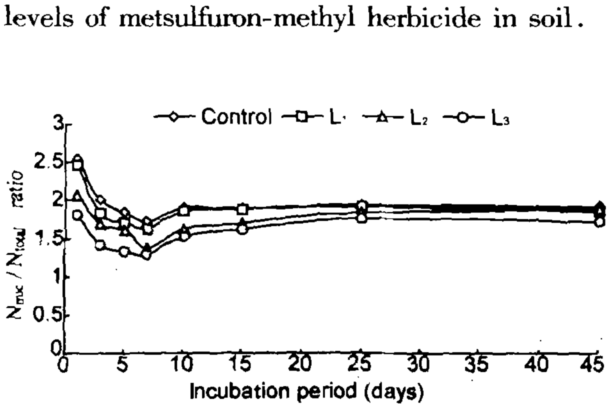

Fig. 4 Effect of metsulfuron-methyl herbicide on microbial biomass N/total N ratio (N_{mic}/N_{total} ratio) in a loamy sand soil

$L_1(0.01\ \mu g \cdot g^{-1})$; $L_2(0.1\ \mu g \cdot g^{-1})$; $L_3(1.0\ \mu g \cdot g^{-1})$

The results indicated that metsulfuron-methyl application at 0.01 $\mu g \cdot g^{-1}$ soil resulted in 3.05%, 8.53%, 7.24%, 5.43%, 2.05%, 1.21%, 1.05%, and 2.87% reduction of N_{mic}/N_{total} after 1, 3, 5, 7, 10, 15, 25, and 45 days of incubation, respectively, compared to the control. At the rate of 0.10 $\mu g \cdot g^{-1}$ soil the reduction was 18.13%, 16.13%, 12.49%, 20.54%, 15.01%, 10.72%, 4.75%, and 2.07%, respectively, relative to the control, with the same periods of incubation. While the addition of 1.00 $\mu g \cdot g^{-1}$ to the soil caused the highest reduction of 28.31%, 30.10%, 27.59%, 25.62%, 20.16%, 15.01%, 8.77%, and 9.95%, respectively, against the control, with the same incubation periods. Fig. 5 curves on the effect of metsulfuron-methyl herbicide on N_{mic}/N_{total} ratio in different incubation periods show that decrease in N_{mic}/N_{total} ratio was in the order 1.00 $\mu g \cdot g^{-1}$ soil > 0.10 $\mu g \cdot g^{-1}$ soil > 0.01 $\mu g \cdot g^{-1}$ soil. These reductions in the ratio markedly increased in the first week after the incubation.

5. The effect of metsulfuron-methyl on (C_{mic}/N_{mic}) ratio

The addition of metsulfuron-methyl to the soil increased the C_{mic}/N_{mic} ratio (Fig. 5). Results indicated that the increase in the C_{mic}/N_{mic} ratio was related to the increased concentrations of herbicide added in the soil. The data indicated that herbicide application at the rate of 0.01 $\mu g \cdot g^{-1}$ soil increased C_{mic}/N_{mic} ratio by 3.5%, 6.9%, 2.4%, 1.2%, 4.8%, 4.8%, 4.7%, and 2.9% after 1, 3, 5, 7, 10, 15, 25, and 45 days of incubation, respectively, as compared with the control. While at 0.10 $\mu g \cdot g^{-1}$ soil herbicide dosage, increased it by 7.2%, 2.5%, 1.6%, 10.6%, 12.1%, 12.9%, 3.9%, and 1.1% at the same incubation periods, respectively, compared with the control. However, at 1.00 $\mu g \cdot g^{-1}$ soil application, it was increased by 16.8%, 13.9%, 13.3%, 10.8%, 8.8%, 11.8%, 4.4% and 10.9% at the same incubation periods, respectively, as compared with the control.

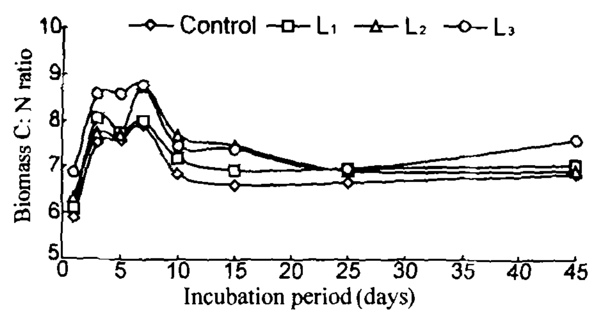

Fig.5 Effect of metsulfuron-methyl on C_{mic}/N_{mic} ratio

$L_1(0.01\ \mu g \cdot g^{-1})$; $L_2(0.1\ \mu g \cdot g^{-1})$; $L_3(1.0\ \mu g \cdot g^{-1})$

DISCUSSION

In our study, soil microbial biomass- C and N were decreased with increasing levels of herbicide applications. This decrease was significant during the first 7 days and 10 days at 0.10 and 1.0 $\mu g \cdot g^{-1}$ soil application rates, respectively. However, at lower rate (0.01 $\mu g \cdot g^{-1}$) the decrease was not significant.

The reason for the decrease of microbial biomass, as affected by metsulfuron-methyl, may be related to the toxicity effect of the herbicide and the adsorption of herbicide in soil, and also to the possibility that the soil microorganisms were not adapted to metsulfuron-methyl. Junnila et al, (1994) reported that metsulfuron persisted longer in sandy soils than in other soils. The phytotoxicity of the 4 g a.i. ha^{-1} dose persisted in the 0 – 5 cm layer of all soils for 1 month on average. The phytotoxicity of the 12 g a.i. ha^{-1} dose persisted for at least 1 month and generally for one year. The fate of sulfonylurea in soils is directly related to their chemical structure and mainly to the ionization of the sulfonylurea bridge (Brown, 1990 ; Walker & Welch, 1989).

Other herbicides such as MCPA and simazine had no detectable effects to the microflora, but repeated paraquat application significantly lowered soil microbial biomass. The results indicated that there may be substantially different effects on soil biomass produced by single or repeated applications of pesticides (Yentum and Johnson, 1986). Perucci and Scarponi (1996) also observed reduced microbial biomass in rimsulfuron herbicide application in a clay loam soil.

The high concentration of metsulfuron-methyl caused a significant decrease in microbial biomass during the early incubation period (first 7days after incubation). This can possibly be explained by the degradation rate, and the half-life of the herbicide. The half life for metsulfuron-methyl ranged from 8 to 36 days (James et al., 1995).

The noted increase of C_{mic}/N_{mic} ratio was related to the decrease of microbial biomass C and N and increase in the levels of metsulfuron-methyl additions. The increase in the level of metsulfuron-methyl herbicide caused subsequent increase in toxicity of the herbicide resulted in greater reduction in N_{mic} in treated soils, as compared with the control. The ratio of biomass C_{mic}/N_{mic} was therefore more in favor of C_{mic} than N_{mic}. Khan and Huang (1998) reported that the increase in biomass C/N ratio can highlight of changes in microbial populations and also suggested that the changes in the biomass C/N could be a good indicator of the changes in the microbial community structure.

The marked decrease in C_{mic}/C_{org} percentage occurred at 0.10 and 1.00 $\mu g \cdot g^{-1}$ soil application, especially within first the 10 to 15 days of incubation because the half-life for metsulfuron-methyl ranged from 8 to 36 days (James et al., 1995). The decrease in the ratio of microbial biomass carbon to total organic carbon (C_{mic}/C_{org}) in herbicide-treated soil may also be attributed to: 1. reduction in the rate of carbon mineralization that, in turn, resulted in an accumulation of organic matter in the soil (Valsecchi et al., 1995); 2. decline in the size of the soil microbial biomass as the result of the toxic effects of the herbicide on microbial populations (Gigliotti et al., 1998).

The decrease in the ratio of the microbial biomass N to total soil N (N_{mic}/N_{total}) percentage in herbicide contaminated soils may be due to the decline in the size of the microbial biomass probably because of the toxic effect of herbicide contamination (Anderson and Barrett, 1985). Walley et al. (1996) suggested that the soil microbial biomass responded rapidly to changes in total C and N associated with site disturbance.

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