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EFFECT OF HERBICIDES ON MICROBIOLOGICAL PROPERTIES OF SOIL

ABSTRACT: Microorganisms decompose herbicides and they may serve as bioindicators of soil changes following herbicide application. Certain microbial species may be used as bioherbicides. This study has shown that *Azotobacter* is most sensitive to herbicide application; it is, therefore, a reliable indicator of the biological value of soil. The numbers of this group of nitrogen-fixing bacteria decrease considerably in the period of 7—14 days after herbicide application. Simultaneously, the numbers of *Actinomycetes* and less so of fungi increase, indicating that these microorganisms use herbicides as sources of biogenous elements. Rate of herbicidal decomposition depends on the properties of the preparation applied, herbicide dose as well as on the physical and chemical soil properties, soil moisture and temperature, ground cover, agrotechnical measures applied and the resident microbial population.

KEY WORDS: microbes, herbicides, bioindicators, inoculants

INTRODUCTION

In modern agricultural production, herbicide application is a regular practice. While in developed countries weeds and pests reduce yields of agricultural crops from 15 to 20%, reductions soar to 50% in undeveloped regions (Dobrovoljskiy and Grishina, 1985). The problems caused by the increased application of herbicides call for multidisciplinary approach. Incorrect and indiscriminate application of herbicides affects negatively the health of humans, plants and animals. Particularly hazardous are the poorly degradable herbicides (triazins) whose persistence may lead to long-term accumulation.

Soil microorganisms are an important link in soil-plant-herbicide-fauna-man relationships. They take part in herbicide a) degradation, their activity, number and diversity may serve as b) bioindicators of changes in soil biological activity following herbicide application and, finally, some microbial species may be used as c) bioherbicides.

Herbicides become incorporated in soil directly, during plant treatment, and indirectly, via water or residues of plant and animal origin. After application, herbicides may evaporate (volatilize), may be washed away through surface run-off, may leach into deep soil strata and ground water, may be inactivated by plants, or may be adsorbed in soil in which case they become subject to chemical or microbiological degradation.

Herbicides are specific regarding their toxic level. However, the application of several chemicals may lead to synergy and development of toxic effects hazardous for humans and the ecosystem (Michaelidou et al., 2000). Herbicides may cause acute and genetic toxicity which are perilous for the biota inhabiting the ecosystem. The halflife of various herbicides ranges from 9 to 116 years. It means that in soil without microorganisms herbicide application would threaten all living things with unforeseeable consequences (Vrochinskiy and Makovskiy, 1979). The European Union has opted for sustainable agriculture, reduced pesticide use and monitoring of acute and genetic toxicity for the ecosystem (Petsikos-Panagiotarou, 2000). Rate of herbicide decomposition in soil depends on the properties of the preparation applied (Mishustin and Emtsev, 1987), herbicide dose (Schuster and Schröder, 1990; Milošević et al., 2001), physical and chemical soil properties (Willems et al., 1996; Miličić, 1987), humidity, temperature, plant cover, soil cultivation technique and the types of the soil microorganisms present (Barriuso and Houot, 1996; Govedarica et al., 1993, 2000; Willems et al., 1996; Milošević et al., 2000a, 2001).

HERBICIDE DEGRADATION BY MICROBES

Herbicide degradation in soil may be photochemical, chemical or microbial in nature. While photochemical decomposition predominates in air and water, only a small percentage of pesticides is decomposed in that way in soil. Chemical decomposition of herbicides in soil evolves through hydrolytic and non-hydrolytic transformations and oxidation. Microorganisms are efficient decomposers of aliphatic and hydroxyl compounds, but they decompose aromatic substances at a slower rate. The compounds that contain oxygen, sulfur or nitrogen in the ring are slowest to decompose (Janjić et al., 1996).

According to Lynch (1983), microbes degrade herbicides in the course of metabolic (when adaptation phenomena take place) and cometabolic processes. New compounds are formed from herbicide metabolites.

In general, herbicides affect microbes indirectly, causing physiological changes, increased enzymatic production or, when applied in high doses, death of susceptible groups of microorganisms (Cervelli et al., 1978). Soil microbiological population uses herbicides and their metabolites as sources of biogenous elements (Cook and Hutter, 1981; Radosevich et al., 1995). It has been noticed that certain groups of microorganisms (primary population) start to decompose herbicides a few days after their arrival. On the other hand, the so-called secondary population, which produces induced enzymes, decomposes herbicides while these are passing through a period of adaptation. Some microbial groups are indifferent to herbicide application (Figure 1). Long-term application (19 years) of glyphosate reduces C biomass in soil, but ammonification and nitrification are increased compared with untreated soil (H a r t and B r o o k e s, 1996).

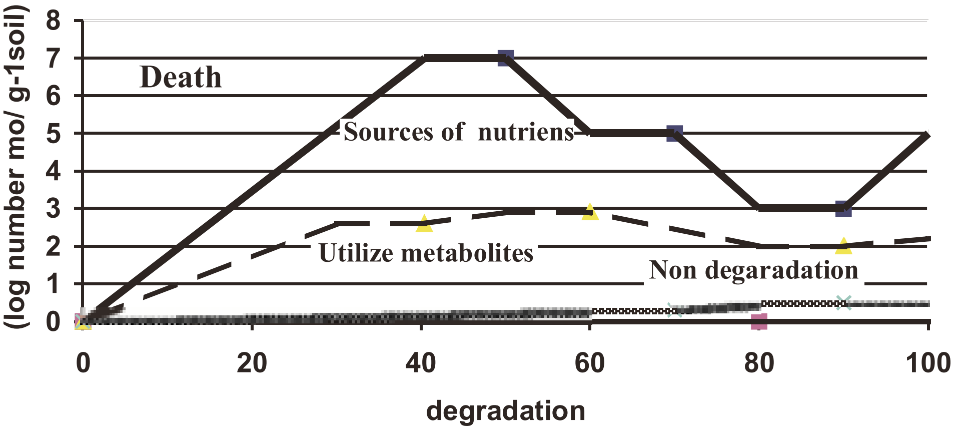


Fig. 1 — Effect of herbicides on soil microbiological population

Studies of numerous authors (L y n c h, 1983; R a d o s e v i c h et al., 1995; M i l o š e v i ć et al., 2001) show that herbicide-decomposing microorganisms belong to bacteria and fungi: *Arthrobacter, Pseudomonas, Bacillus, Actinomycetes, Mycoplana, Agrobacterium, Corynebacterium, Arthrobacter, Flavobacterium, Nocardia* and *Trichoderma*. Effect of herbicides on the composition and morphology of soil microbial population depends on the composition and dose of herbicides applied but also on the kind of microorganisms present (M i š k o v i ć et al., 1983; M i l o š e v i ć et al., 2001). In general, herbicides affect soil microbes indirectly. Herbicides may be a source of nutrition for microbes (C o o k and H u t t e r, 1981), in which case they significantly affect microbial growth and multiplication. However, herbicides also affect the microbes physiologically: a) by changing their biosynthetic mechanism (a change in the level of protein biosynthesis is reflected on the ratio of extracellular and intracellular enzymes); b) by affecting protein biosynthesis (induction or repression of synthesis of certain enzymes); c) by affecting the cellular membranes (changes in transport and excretion processes); d) by affecting plant growth regulators (transport of indolacetic acid, gibberellin synthesis and ethylene level); e) applied in high doses, they may kill microorganisms.

FACTORS AFFECTING MICROBIOLOGICAL DECOMPOSITION OF HERBICIDES

Rate of herbicide decomposition in the soil is influenced by the properties of the preparation applied, its dose as well as by the physical and chemical soil properties, soil moisture and temperature, plant cover, soil cultivation method and kinds of microorganisms present (Miličić, 1987; Schuster and Schröder, 1990; Radosevich et al., 1995; Milošević et al., 2001).

Herbicide properties

According to their structure, herbicides may be carbonic acids and their derivatives, aryloxyalkyl carbonic acids and their derivatives, carbamic acid derivatives, carbamide derivatives, thio- and dithiocarbamic acid derivatives, dipyridil derivatives, nitrophenols, nitroanilines or heterocyclic compounds comprising nitrogen in the ring (diazin and triazin). Toxicity, persistence and selectivity are important characteristics of herbicides (Konstantinović et al., 1998, 1999).

Table 1 — Period of herbicide degradation in soil (Konstantinović, 1999)

1—3 months	3—6 months	more than 12 months
Aminotriazol	Butan	Atrazine
Aziprotryne	Chlorbromuron	Bromacil
Carbetamide	Chloridazone	Chlortiamid
Chlorpropham	Chlortal-dimethyl	Dichlorbenil
Cyanazine	Chlortoluron	Lenacil
Dalapone	Cikloat	Methazone
Prometryn	Dinitramin	Metribuzin
Propahlor	EPTC	Napropamid
Propham	Etophumesat	Oksadiazon
Terbutrin	Izoproturone	Fenmedipham
2,4-D	Linuron	Pikloram
MCPA	Methamitron	Propyzamide
Dichloprop	Matolachlor	Simazine
	Mathobentiazuron	Terbacil
	Metabromuron	Trifluralin
	TCA	Imidazolinone
	Trietazin	Sulfonylurea

Regarding their degradation period, pesticides may be divided in two groups: a) residual, with long toxic action and b) contact, with short toxic action. A study of Barriuso and Houot (1996) showed that simazine mineralizes faster than atrazine. A hormone herbicide 2,4-D decomposes in soil very fast. It is decomposed by several microorganisms: *Mycoplana, Corynebacterium, Achromobacter, Rhizobium, Arthrobacter, Flavobacter* and some actinomycetes (Lynch, 1983).

Herbicides dose

Increase in herbicide dose tends to amplify its negative effect on microorganisms. High herbicide concentrations reduce the number of nodules in symbiotic nitrogen-fixing microoorganisms, nitrogenase activity microorganisms, dry matter in plants, lysis of bacteroids, and inhibition of ATP synthesis (Govedarica et al., 1993; Konstantinović et al., 1998; Milošević et al., 2001).

A dose of 1.6 l. ha^{-1} of dimethenamide (Frontier) caused larger reductions of the total number of microorganisms and azotobacters by 5—7% and 2—18%, respectively, than a dose of 1.4 l. ha^{-1} (Milošević et al., 2001a). A dose of 2 l. ha^{-1} of flumetsulam + trifluralin (Rival) caused a larger reductions of *Azotobacter* by about 2% than a dose of 1.7 l. ha^{-1} (up to 30 days) (Milošević et al., 2000a; Govedarica et al., 2001). Simultaneously, the larger dose of Rival increased the numbers of fungi and actinomycetes by 2—4% and 1%, respectively (Milošević et al., 2001a). Increased doses of dimethenamid (1.6 l. ha^{-1}) and metolachlor (1.7 l. ha^{-1}) caused larger reductions in the number of azotobacters than lower doses (Govedarica et al., 2001).

High doses of atrazine and alachlor (3 and 4 l·ha^{-1}, respectively) caused decreases in the total number of bacteria, ammonifiers and azotobacters and they reduced dehydrogenase activity (Konstantinović et al., 1999).

Under laboratory conditions, a normal dose of glyphosate inhibited DHA by 5—10% (3 weeks after herbicide application). A tenfold dose of glyphosate affected negatively the activity of this oxide-reducing enzyme by 5% (11 weeks after herbicide application) (Schuster and Schröder, 1990).

Physical and chemical soil properties

Pesticide adsorption or desorption depends on the physical and chemical soil properties. The process of adsorption depends on the concentration and solubility of herbicides in soil solution, ion exchange capacity, organic matter content, pH, moisture and temperature of soil, etc. Soils with heavy mechanical composition have a higher pesticide-adsorbing capacity than light (sandy) soil. Studies have offered different results regarding the rate of atrazine mineralization in soil. Atrazine mineralization is exceedingly low, which is an indication that this pesticide is very persistent. According to Willems et al. (1996), atrazine mineralization is slow and controlled by the amount of biomass and organic C, and it decreases with depth. Atrazine mineralization seems to be predominantly due to cometabolism rather than direct metabolism. Wolf and Martin (1975) stated that atrazine mineralization was 18% after 550 days, depending on physical and chemical soil properties. Klint et al. (1993) reported the value of 20% after 90 days. According to Willems et al. (1996), the amounts of 2,4-D mineralized in the soil layer between 1m and 1.5 m were generally high, exceeding on average four times the amounts mineralized in the topsoil. The authors hypothesized that the rapid mineralization in deeper soil layers was probably due to complex interactions among microbial activity, microbial population structure, nutrition status and the physical and chemical properties of the soil.

Temperature and moisture

Increased moisture and temperature accelerate the degradation of atrazine and 2,4-D (W i l l e m s et al., 1996). The rate of atrazine mineralization is slow (< 2%), further decreasing in deeper soil layers, at low temperature and at low moisture content. Over a period of two and a half months, 10 to 80% of 2,4-D were mineralized, depending on the depth of soil profile.

Cultural practices

In well-tilled and loose soil that contains much oxygen, the rate of herbicide degradation is accelerated because of dynamic microbiological processes taking place in it. In uncultivated or excessively compacted soil, where anaerobic processes predominate, the rate of herbicide degradation is low. Methods of soil cultivation and herbicide application affected significantly the microbiological activity (M i l o š e v i ć et al., 1995, 1995a). Generally, chiseling and rototilling following glyphosate application tended to increase the numbers of soil microorganisms. Glyphosate application in bands reduced the number and enzymatic activity of soil microorganisms in relation to the broadcast application.

Application of NPK fertilizers reduced the degradation of the hormone herbicides 2,4-D and MCPA by 30—50%. Application of $CaCO_3$ considerably lowered the rate of herbicide degradation. No degradation could be registered 14 days after the application; on the 28^{th} day, 2,4-D of MCPA were degraded by 29% and 45%, respectively (B u r n s , 1995).

Negligent use of heavy machines for soil tillage and sugarbeet transport under unfavorable weather conditions (rain and high soil moisture) may cause extensive soil structure deterioration and prolonged negative effects on subsequent crops (M i l o š e v i ć et al., 2001). Under unfavorable, i.e., anaerobic conditions, soil microorganisms produce metabolites which, synergizing with the applied herbicides, intensify their negative effects (K o s i n k i e v i c z, 1984a). Bacteria from the genus *Arthrobacter* sp., isolated from sugarbeet rhizosphere, produced phenolic compounds when exposed to soil and anaerobic conditions. These compounds intensified the phytotoxic effect of cycloate (Roneet) on the crop planted after sugarbeet (K o s i n k i e v i c z et al., 1984).

Plant cover

Plants play a role in pesticide decomposition. They incorporate pesticide via roots, stems and leaves. Pesticide dose and mode of application determine how it is incorporated, by the aboveground plant parts or the roots (Kastori et al., 1996). Plant species affects the rate of mineralization of atrazine's triazine rings. Experiments of Barriuso and Houot (1996) showed that atrazine's triazine rings were mineralized faster in the soils under corn treated with this herbicide each year that in the untreated soils under wheat or grasses.

Milošević and Govedarica (2001) noted that prometryne application reduced some parameters of biological activity (total number of bacteria and the number of azotobacters) in the soil under soybean and sunflower. In the soil under soybean and sunflower, however, azotobacters could not be found 28 days after prometryne application while 45 days after the application its numbers reached up to 10.76 x 10^2 per one gram of soil.

Microbiological population

Pesticide decomposition is affected by the size and composition of microbiological population. Microorganisms are a highly heterogeneous group, including aerobes and anaerobes, heterotrophs and autotrophs or saprophytes, symbionts and parasites. Certain microbial species may decrease or increase the toxic action of herbicides. Most soil microorganisms are capable of decomposing herbicides, using them most frequently as sources of biogenous elements. Experiments have shown that microbes may use atrazine as a source of carbon (Radosevich et al., 1995) or nitrogen (Cook and Hutter, 1981). The hormone herbicide 2.4-D is rapidly decomposed in the soil, as much as 3.4 x 10^6 moles in a single day. Various microorganisms decompose 2.4-D: *Mycoplana, Corynebacterium, Achromobacter, Rhizobium, Arthrobacter, Flavobacter* and some actinomycetes (Lynch, 1983). Herbicides derived from the carbamic acid (phenmedipham, desmedipham) are decomposed microbiologically, chemically and photochemically. Among the fungi, *Rhizopus japanus, Aspergillus* ssp., *Penicillium* ssp. and *Metharizium anosoplie* are the most intensive decomposers. Among the bacteria, those are *Pseudomonas* sp. and *Bacillus* sp. According to Kosinkievicz (1984a), the phenolic compounds produced by *Pseudomonas acidovorans* may increase or decrease the phytotoxicity of lenacil (Venzar), the mode of action depending on herbicide concentration.

MICROBES: BIOINDICATORS AND INOCULANTS

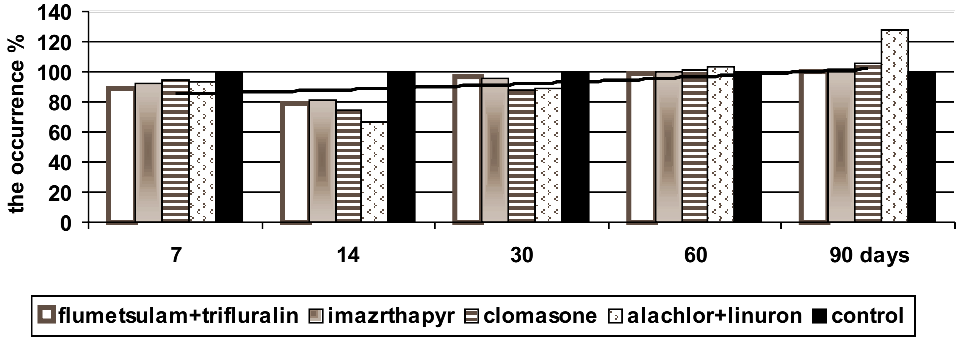
The soil is a highly complex system. It is also dynamic, on account of microorganisms whose enzymes take part in most syntheses and decompositions. The number, enzymatic activity and biodiversity of microorganisms may serve as indicators of soil fertility, as well as indicators of all changes taking place in the soil as an ecological system (Milošević et al., 1997).

By means of adsorption, bioaccumulation and the production of metabolites, microorganisms are capable of mitigating or blocking the toxic action of herbicides (Janjić et al., 1996). Herbicides adsorb on cell surface, affecting ion transport. They affect the metabolism inside the cell by binding to amino and sulfide groups. In the course of these processes, changes take place in the oxidoreduction level of soil and, depending on the chemical composition and dose of herbicide, the microorganism concerned may be killed. Bioaccumulation mitigates the toxic effect of herbicides.

Bioindicators

Application of pesticides and other chemicals used in agriculture affects the vital functions and population dynamics of soil microorganisms. Microorganisms are a heterogeneous group of organisms whose enzymatic systems comprise 60—90% of the total metabolic activity of the soil (L e e , 1994). Population size, enzymatic activity and biodiversity of certain systematic and physiological groups of microorganisms may serve as bioindicators of changes taking place in the soil following herbicide application (M i l o š e v i ć et al., 1995, 2001; G o v e d a r i c a et al., 1993, 1995; K o n s t a n t i n o v i ć et al., 1999). Results of our studies have shown that generally, herbicides tended to reduce the total number of soil microorganisms 7 to 30 days after application (M i l o š e v i ć et al., 2001).

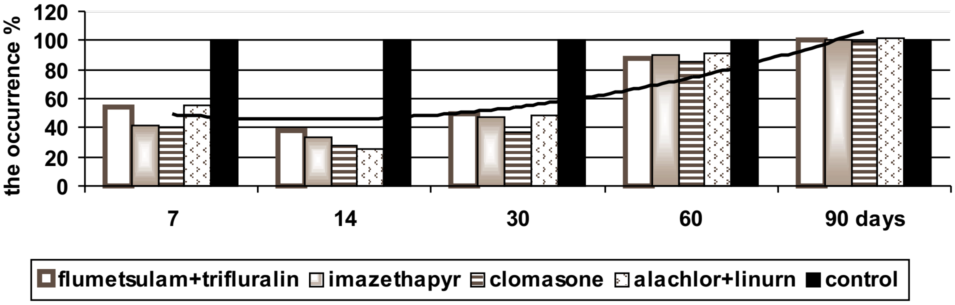
M i l o š e v i ć et al. (1998, 2000a) reported that soybean treatment with flumetsulam + trifluralin (Rival), imazethapyr (Pivot), clomazone (Command) and alachlor + linuron (Linuron S-50) reduced the total number of microorganisms in the period of 14 days after application by 15 to 27% (Graph 1). After that period, the numbers of microorganisms in the treated variants reached the level of the control variant while on the 90th day after application the numbers of microorganisms in the variants with clomazone (Command) and alachlor + linuron (Linuron S-50) were increased in relation to the control (M i l o š e v i ć et al., 2000a).



Graph. 1 — Effect of herbicides on the total number of microorganisms

Nitrogen-fixing bacteria are important from the point of nitrogen balance in the soil and their reaction to herbicides is a good indicator of how effective an applied weed control action is.

Our studies have shown that *Azotobacter* is most sensitive to herbicide application (M i l o š e v i ć et al., 1995; 2000a, M i l o š e v i ć and G o v e - d a r i c a , 2000), and it may serve as a reliable indicator of the biological value of soil. Herbicide application reduced the number of azotobacters (Graph 2). The reduction was large in the period of 30 days after herbicide application. The largest rates of reduction occurred 14 days after application, 62% in the case of Rival and 78% in the case of Linuron S-50.



Graph. 2 — The effect of herbicides on the occurrence of *Azotobacter*

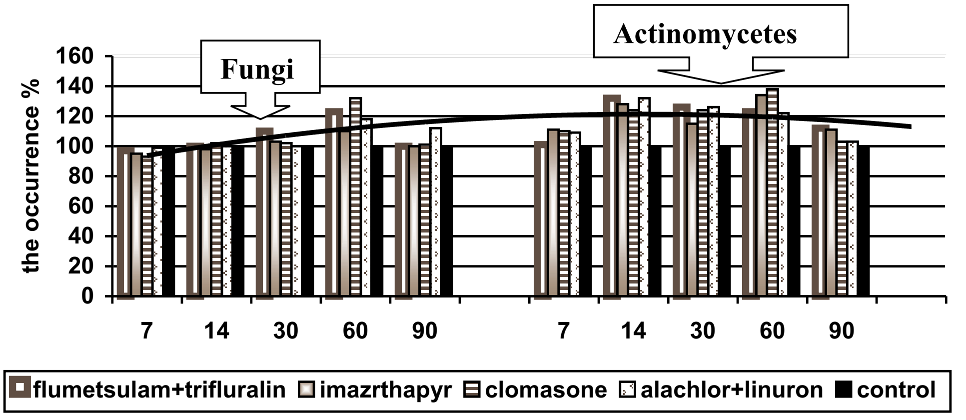
M i l o š e v i ć and G o v e d a r i c a (2001) found that the negative effect of prometryne on the growth and development of azotobacters was higher in the soil under sunflower than in the soil under soybean. In the sunflower field, azotobacters could not be registered in the soil 28 days after prometryne application, but on the 45^{th} day their number reached 10.76×10^2 per one gram of soil. According to G o v e d a r i c a et al. (2001), the number of azotobacters was 2—3 times lower in a sugarbeet field treated with dimethenamide and metalochlor than in the untreated control variant.

Herbicide application also inhibits the symbiotic bacterium *Bradyrhizobium japonicum.* Nodulation rate is an indicator of symbiotic efficiency between soybean plant and the bacterium. Nodulation test shows the efficiency of nitrogen utilization in the process of biological nitrogen fixation. Herbicide application reduced the nodulation rate, i.e., the number of nodules formed on soybean roots, by 5—21% (M i l o š e v i ć et al., 2000a).

According to C e r v e l l i et al., (1978), herbicides may kill sensitive microorganisms. However, herbicides may also be decomposed by enzymes produced by microorganisms, which subsequently use the metabolites as sources of biogenous elements (C e r v e l l i et al., 1978; M i l o š e v i ć et al., 2001). It has been noticed that certain groups of microorganisms (the primary population) start to decompose herbicides a few days after their applications. However, the secondary population, which produces induced enzymes, starts to decompose herbicides after a period of adaptation.

Low concentrations of 2,4-dichlor-phenoxy-acetic-acid (2,4-D) promote the development of tumorous structures (p-nodules) on the roots of corn, wheat (C h r i s t a i n s e n - W e n i g e r , 1992; 1995; C h r i s t a i n s e n - W e - n i g e r and V a n d e r l e y d e n , 1994, cit. C h r i s t a i n s e n - W e n i g e r , 1997) and rice (C h r i s t a i n s e n - W e n i g e r , 1997). On inoculation, *Azo-*

spirillum brasilinse colonizes *p*-nodules. The nitrogenase activity of tumors inhabited by bacteria is significantly increased in comparison with the untreated control.

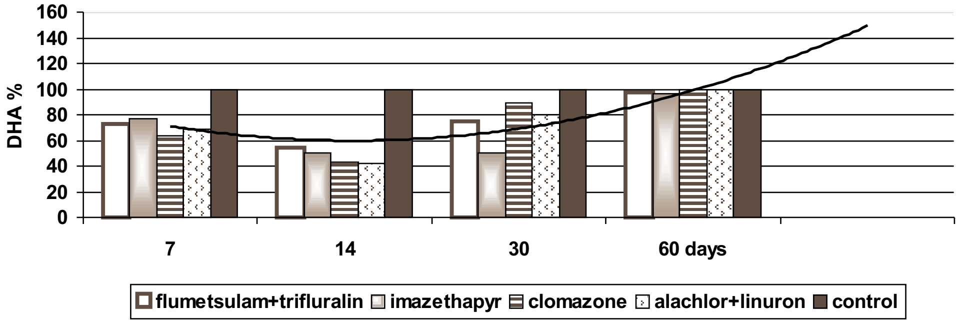


Graph. 3 — Effect of herbicides on fungi and actinomycetes

Generally, our studies have shown that herbicide application stimulated the growth and development of actinomycetes. Dimethenamide and metalochlor applied to sugarbeet (G o v e d a r i c a et al., 2001), and prometryn applied to soybean and sunflower (M i l o š e v i ć and G o v e d a r i c a, 2001) increased the number of actinomycetes. Only seven days after herbicide application, their numbers started to increase (Graph. 3). On the 14th day, the numbers of actinomycetes were increased by 35% in the treatments with Rival (flumetsulam+ trifluralin) and Linuron (alachlor+linuron). The application of imazethapyr and clomazone stimulated the development of actinomycetes, their number reaching the maximum value 60 days after herbicide application. The effect of herbicides on fungi is variable and it depends on the dose applied (M i l o š e v i ć et al., 2000; M i l o š e v i ć and G o v e d a r i c a, 2001).

Enzymes are catalysts of biochemical processes in the soil and they take part in the cycling of carbon, nitrogen, phosphorus and sulfur. They play an important role in the maintenance of soil fertility and their activity may serve for the assessment of soil fertility (S k u j i n š, 1973; G o v e d a r i c a et al., 1993; M i l o š e v i ć et al., 1997).

Being endocellular, oxido-reducing enzymes, dehydrogenases are a foundation of the enzymatic systems of all microorganisms (v o n M e r s i and S c h i n n e r, 1991) and their role is essential in initiating the oxidation of organic matter in the soil, by transporting electrons or hydrogen from the substrate to the sink (R o s s, 1971). DHA is a measure of microbial oxidative activity (S k u j i n š, 1973; C a s i d a, 1977; T a b a t a b a i, 1982; T r e v o r s, 1984; C a m i ñ a et al., 1998), Many authors consider it as an indicator of soil biological activity (L e n h a r d, 1956; T h a l m a n n, 1968; S k u j i n š, 1973; M i l o š e v i ć et al., 1993, 1996). Our previous studies showed that DHA is a good indicator of changes in oxido-reducing processes in the soil taking place in consequence to herbicide application (M i l o š e v i ć et al., 1995, 1998, 2001; G o v e d a r i c a et al., 1995, 2001).



Graph. 4 — Effect of herbicides on dehydrogenase activity

The tested herbicides (Graph 4) were significantly reducing dehydrogenase activity on the 7th, 14th and 30th day after application (M i l o š e v i ć et al., 2001). After that period, however, the oxido-reducing processes return to the level of the control variant.

According to G o v e d a r i c a et al. (2001) metalochlor caused a larger reduction in dehydrogenase activity than dimethenamide. The application of a combination dimethenamide + chloridazon inhibited the oxido-reducing processes in the soil to a lesser extent than the application of individual herbicides. The values of DHA were on the level of the control variant 180 days after herbicide application. DHA was reduced after the application of prometryn (M i l o š e v i ć and G o v e d a r i c a, 2001), but it was back on the level of the control variant 45 days after application.

DHA is a sensitive indicator of side-effects of herbicide application on non-target soil microorganisms (S c h u s t e r et al., 1987, cit. to S c h u s t e r and S c h r ö d e r, 1990a). A field trial of S c h u s t e r and S c h r ö d e r (1990a) showed a decreased dehydrogenase activity following the application of dichlorprop (13%) and glyphosate (5%). Under laboratory conditions, a normal dose of glyphosate stimulated DHA during the period of six weeks after application. A ten-fold dose of glyphosate, however, reduced the oxido-reducing processes by 5% to 25% (S c h u s t e r and S c h r ö d e r, 1990).

According to J a n j i ć et al. (1996) the amount of released CO_2 (as a measure of the intensity of soil microbiological activity) was reduced after the application of atrazine and cyanazine. Repeated atrazine applications caused a significant reduction in the intensity of soil respiration followed via the amount of released CO_2 (D ž u g e l i, 1982., cit. J a n j i ć et al., 1996).

Inoculants

Microorganisms may be added to soils contaminated with herbicides in order to mitigate negative residual effects on subsequent crops. Examining 16 herbicides, B u r n s (1995) selected two hormonal herbicides as indicators of microbiological degradation: 2(2,4-dichlorophenoxy) acetic acid (2,4-D) and 2-methyl-4-chlorphenoxyacetic acid (MCPA). Fourteen days after soil inoculation with bacteria isolated from contaminated soils, the rates of herbicide decomposition were 67—78% in the case of 2.4-D and 84—98% in the case of MCPA. The rate of decomposition depended on bacterial strain used for inoculation (B u r n s , 1995). In the non-inoculated soil, the rate of decomposition was 23—27%. However, 28 days after inoculation, the amounts of decomposed herbicides in the treated and untreated soils were equal. MCPA was decomposed faster that 2,4-D, 99% of it being decomposed 28 days after inoculation.

Weeds may be successfully controlled with preparations based on fungi (*Beauveria bassiana, Paeciloyces litacinus, Verticillum chlamydosporum*). It was found that *Sphacelotheca halepense* may be used as a mycoherbicide in the biocontrol of *Sorgum halepense* (M i l a n o v a and K a r a đ o v a , 1997).

CONCLUSION

Microorganisms are constituting elements of the environment. Their abundance, enzymatic activity and biodiversity are good indicators of the balance in the agro-ecological system. To use microorganisms as indicators of changes in soil biological activity, it is necessary to find most reliable detection methods and most effective microbial strains. Inoculants based on certain microorganisms may be used as biopreparations in combination with or instead of chemical preparations. They are applicable in integrated weed management, which implies combining of cultural practices, biological and chemical methods of weed control. Microbiological research should be focused on isolating microbial strains which, on application, effectively decompose herbicides, irrespectively of the natural microbial population in the soil.

It is necessary to keep strengthening the scientific basis of modern agriculture because herbicides may be advantageously used only if their persistence, bioaccumulation and toxicity (acute and genetic) in agro-ecosystems are strictly controlled.

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УТИЦАЈ ХЕРБИЦИДА НА МИКРОБИОЛОШКА СВОЈСТВА ЗЕМЉИШТА

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Резиме

Микроорганизми земљишта су значајна карика у односу земљиште — биљка — хербициди — фауна/човек. Наиме, микроорганизми а) учествују у деградацији хербицида, б) њихова активност, бројност и разноврсност су биоиндикатори промена биолошке активности земљишта после примене ових хемијских једињења, и в) поједине врсте микроорганизама могу се применити као биохербициди.

Хербициди изазивају угинуће осетљивих група микроорганизама. Међутим, хербициди се могу разградити у земљишту ензимима микробних популација користећи ова хемијска једињења и добијене метаболите као изворе биогених елемената. Уочава се да поједине групе микроорганизама (примарна популација) разграђују хербициде већ после неколико дана након доспевања у земљиште. Међутим, тзв. секундарна популација која продукује подесне-индуковане ензиме разграђује ова хемијска једињења после периода адаптације.

Наша истраживања показују да је група микроба *Azotobacter* најосетљивија на примену хербицида, те може бити поуздан биоиндикатор биогености земљишта. Уочава се да је заступљеност ове групе азотофиксатора смањена у великом

проценту до 14. дана. Заступљеност *Actinomycetes* и у мањој мери гљива је повећана, што указује да ове групе микроорганизама користе хербициде као изворе биогених елемената.

Разградња хербицида у земљишту зависи од својства примењеног препарата, затим од количине (дозе) хербицида, али и од физичко-хемијских својстава земљишта (влажности, температуре, биљног покривача, обраде) и врсте микроорганизама.