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Degradation of glyphosate and other pesticides by ligninolytic enzymes

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Abstract The ability of pure manganese peroxidase (MnP), laccase, lignin peroxidase (LiP) and horseradish peroxidase (HRP) to degrade the widely used herbicide glyphosate and other pesticides was studied in separate in vitro assays with addition of different mediators. Complete degradation of glyphosate was obtained with MnP, $MnSO_4$ and Tween 80, with or without H_2O_2. In the presence of $MnSO_4$, with or without H_2O_2, MnP also transformed the herbicide, but to a lower rate. Laccase degraded glyphosate in the presence of (a) 2,2′-azino-bis(3-ethylbenzthiazoline-6-sulphonic acid) (ABTS), (b) $MnSO_4$ and Tween 80 and (c) ABTS, $MnSO_4$ and Tween 80. The metabolite AMPA was detected in all cases where degradation of glyphosate occurred and was not degraded. The LiP was tested alone or with $MnSO_4$, Tween 80, veratryl alcohol or H_2O_2 and in the HRP assay the enzyme was added alone or with H_2O_2 in the reaction mixture. However, these enzymes did not degrade glyphosate. Further experiments using MnP together with $MnSO_4$ and Tween 80 showed that the enzyme was also able to degrade glyphosate in its commercial formulation Roundup® Bio. The same enzyme mixture was tested for degradation of 22 other pesticides and degradation products present in a mixture and all the compounds were transformed, with degradation percentages ranging between 20 and 100%. Our results highlight the potential of ligninolytic enzymes to degrade pesticides. Moreover, they suggest that the formation of AMPA, the main metabolite of glyphosate degradation found in soils, can be a result of the activity of lignin-degrading enzymes.

Keywords Glyphosate · Manganese peroxidase · Laccase · Ligninolytic enzymes · Herbicide · Pesticides

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Introduction

It is generally considered that microbial metabolic and cometabolic processes are the most significant processes for pesticide degradation in the environment (Torstensson 1987), but more information is needed to identify the sources of the often high degree of variability found in pesticide degradation rates (Sims and Cupples 1999).

Peroxidases and laccases, mainly produced by white rot fungi (Kirk and Farrell 1987), are key enzymes involved in lignin degradation (Boerjan et al. 2003). Since these enzymes are non-specific,

they can simultaneously attack a wide range of other organic compounds, and this characteristic has been exploited for bioremediation purposes (Barr and Aust 1994) of, for instance, PAH, chlorinated compounds and dyes (Kotterman et al. 1998; Paszczynski and Crawford 1995; Wesenberg et al. 2003). Furthermore these enzymes can be produced at large scale (Herpoël et al. 1999; Nüske et al. 2002) and can be purified and used for cell-free enzymatic transformation of pollutants (Gianfreda and Rao 2004).

Fungal lignin-degrading systems can also transform pesticides such as bentazon, isoproturon, metribuzin and methoxychlor (Castillo et al. 2000, 2001, Castillo and Torstensson 2007; Hirai et al. 2004) and pesticide metabolites, e.g., 3,5,6-trichloro-2-pyridinol (TCP), a product of chlorpyrifos degradation (Coppola et al. 2007). This degradation ability has been the basis for the design of biobeds, a system originating in Sweden that minimises contamination during the handling of concentrated pesticides before application and by their diluted residues on and in equipment after spraying (Castillo et al. 2008). The principle is that spills of pesticides are collected on the biobed, which contains a mixture of straw, soil and peat. In this biomixture, the growth of white rot fungi is favoured and the production of ligninolytic enzymes is promoted, which correlates with the degradation of pesticides (Castillo and Torstensson 2007).

Plant peroxidases have also been studied in biodegradation of pollutants: horseradish peroxidase has been shown to catalyse the degradation of azo dyes (Bhunia et al. 2001; Mohan et al. 2005), while peroxidases from the roots of shepherd's purse (Park et al. 2006) and turnip (Duarte-Vázquez et al. 2003) have been shown to degrade phenolic compounds.

The transformation of some organic compounds by these enzymes is often enhanced, and in some cases only possible, in the presence of mediators such as veratryl alcohol (Faison et al. 1986), ABTS (2,2'-azino-bis(3-ethylbenzthiazoline-6-sulphonic acid)) (Bourbonnais and Paice 1990), HBT (1-hydroxybenzotriazole) (Böhmer et al. 1998; Kapich et al. 1999), manganese (Wariishi et al. 1992) or lipid radicals (Kapich et al. 1999).

Based on these previous experiences and in order to better understand the possible contribution of peroxidases and laccases to pesticide degradation in the environment, we studied the ability of these enzymes to also degrade glyphosate, the most used

herbicide in the world, and other pesticides. In vitro degradation tests were conducted using purified laccase, manganese peroxidase (MnP), lignin peroxidase (LiP) and horseradish peroxidase (HRP), in combination with different mediators. In addition, the ability of purified MnP to transform glyphosate in its commercial formulation (Roundup® Bio), to degrade the glyphosate metabolite AMPA and to degrade pesticides in a mixture was further tested, also under in vitro conditions.

Materials and methods

Enzymes

Horseradish peroxidase (EC 1.11.1.7), lignin peroxidase (EC 1.11.1.14) and laccase from *Trametes versicolor* (1.10.3.2) were purchased from Sigma–Aldrich (Steinheim, Germany). Manganese peroxidase (EC 1.11.1.13) from *Nematoloma frowardii* was obtained from JenaBios (Jena, Germany).

Chemicals

Glyphosate [*N*-(phosphomethyl)glycine], AMPA [(aminomethyl)phosphonic acid] and the Pesticide Mix 34 were supplied by Dr. Ehrenstorfer GmbH (Augsburg, Germany). Tween® 80 was supplied by Merck (Darmstadt, Germany). Veratryl alcohol (3,4-dimethoxybenzyl alcohol), ABTS (2,2'-azino-bis(3-ethylbenzthiazoline-6-sulphonic acid)), TFAA (trifluoroacetic anhydride ReagentPlus®) and TFE (2,2,2-trifluoroethanol ReagentPlus®) were supplied by Sigma–Aldrich (Stockholm, Sweden). The labelled glyphosate (2-13C, 99%; ^{15}N, 98%) and AMPA (^{13}C, 99%; ^{15}N, 98%; methylene-D2, 98%) internal standards were supplied by LGC Standards (Borås, Sweden). Roundup® Bio was a gift from Monsanto, Sweden.

Degradation of glyphosate by peroxidases and laccases

The potential of laccase, manganese peroxidase (MnP), lignin peroxidase (LiP) and horseradish peroxidase (HRP) to degrade glyphosate was evaluated in separate in vitro assays. The experimental

setup was the same for all enzymes and only the composition of the mixtures changed. Buffers were autoclaved and reagents were sterilised by filtration through 0.45-μm pore size filters before use. The enzymes, or H_2O_2 when present, were added last into the reaction mixtures to start the reactions. All reactions were conducted in sterile loosely capped 8-ml glass vials. The vials were placed on a rotary shaker (150 rpm) at 35°C and samples were taken after 24 h. On each sampling occasion, the samples (50 μl) were mixed with methanol (200 μl) to stop the enzymatic reaction (Eibes et al. 2005) and kept at −20°C until analysis.

In the laccase assay, the standard reaction mixture (1 ml) contained 0.06 mM glyphosate and pure enzyme (0.15 U ml^{-1}) in 100 mM phosphate buffer (pH 6.0). The effect of $MnSO_4$, Tween 80 and ABTS, separately and in combination, was tested. The composition of the reaction mixture for each of the treatments is shown in Table 1.

In the MnP assay, the standard reaction mixture (1 ml) contained 0.06 mM glyphosate and pure enzyme (0.15 U ml^{-1}) in 50 mM Na acetate buffer (pH 4.5). The effect of $MnSO_4$, Tween 80 and H_2O_2, separately and in combination, was tested. The composition of the reaction mixture for each of the treatments is shown in Table 2.

In the LiP assay, the standard reaction mixture (1 ml) contained 0.06 mM glyphosate, pure enzyme (0.15 U ml^{-1}) and 0.05 mM H_2O_2 in 100 mM Na tartrate buffer (pH 3.0). The effect of $MnSO_4$, Tween 80 and veratryl alcohol was tested.

In the HRP assay, the standard reaction mixture (1 ml) contained 0.06 mM glyphosate, pure enzyme (0.15 U ml^{-1}) and 0.2 mM H_2O_2 in 100 mM phosphate buffer (pH 5.0).

In all the assays, the degradation percentages for each treatment were calculated based on the concentration of glyphosate in a control with the corresponding buffer and 0.06 mM glyphosate.

Table 1 Final reagent concentration in the reaction mixtures and glyphosate degradation by laccase under in vitro conditions after 24 h of incubation at 35°C and 150 rpm

	Laccase (0.15 U ml^{-1})	Phosphate buffer (100 mM pH 6.0)	Glyphosate (0.06 mM)	$MnSO_4$ (1 mM)	ABTS (1 mM)	Tween 80 (1% v/w)	Glyphosate degradation (%)
Lac 1	+	+	+	−	−	−	0.0 ± 4.2
Lac 2	+	+	+	−	+	−	40.9 ± 3.6
Lac 3	+	+	+	+	−	+	62.8 ± 0.7
Lac 4	+	+	+	+	+	+	90.1 ± 0.3

Degradation values are means ± SD ($n = 2$)

Table 2 Final reagent concentration in the reaction mixtures and glyphosate degradation by MnP under in vitro conditions after 24 h of incubation at 35°C and 150 rpm

	MnP (0.15 U ml^{-1})	Na acetate buffer (50 mM pH 4.5)	Glyphosate (0.06 mM)	$MnSO_4$ (1 mM)	H_2O_2 (0.05 mM)	Tween 80 (1% v/w)	Glyphosate degradation (%)
M 1	+	+	+	−	−	−	1.6 ± 0.0
M 2	+	+	+	−	+	−	0.0 ± 0.8
M 3	+	+	+	+	−	−	53.0 ± 10.8
M 4	+	+	+	+	+	−	77.6 ± 3.9
M 5	+	+	+	−	−	+	8.7 ± 0.8
M 6	+	+	+	−	+	+	3.3 ± 0.8
M 7	+	+	+	+	−	+	100 ± 0.0
M 8	+	+	+	+	+	+	100 ± 0.0

Values are means ± SD ($n = 2$)

Degradation of formulated glyphosate by MnP

The potential of MnP to degrade formulated glyphosate (Roundup® Bio) was evaluated in vitro. All reactions were conducted in sterile, loosely capped, 8-ml glass vials. The vials were placed on a rotary shaker (150 rpm) at 35°C and samples were taken at the beginning of the experiments and after 1, 4 and 7 days. On each sampling occasion, the samples (50 μl) were mixed with methanol (200 μl) to stop the enzymatic reaction (Eibes et al. 2005) and were kept at −20°C until analysis. The standard reaction mixture (1 ml) contained Roundup® Bio (0.06 mM glyphosate), 1 mM $MnSO_4$, 1% v/w Tween 80 and pure enzyme (0.15 U ml^{-1}) in 50 mM Na acetate buffer (pH 4.5). A control consisting of the standard reaction mixture but without the enzyme was included.

Degradation of the metabolite AMPA by MnP

The potential of MnP to degrade the metabolite AMPA was evaluated in vitro. The experimental setup was the same as for the glyphosate degradation studies. The standard reaction mixture (1 ml) contained 0.06 mM AMPA, 1 mM $MnSO_4$, 1% v/w Tween 80 and pure enzyme (0.15 U ml^{-1}) in 50 mM Na acetate buffer (pH 4.5). A parallel with the standard reaction mixture but without the enzyme was run as a control.

Degradation of a pesticide mixture by MnP

The potential of MnP to degrade pesticides and pesticide degradation products in a mixture (Pesticide Mix 34) was evaluated in vitro. The standard reaction mixture (2 ml) contained pure enzyme (1.5 U ml^{-1}), 1 mM $MnSO_4$, 1% v/w Tween 80 and 0.05 mM H_2O_2 in 50 mM Na acetate buffer (pH 4.5) and the mixture of the pesticides (0.45 μg ml^{-1} each).

The reaction was started by addition of MnP and additional 0.15 units of enzyme were added daily after sampling for a total reaction time of 6 days. A control consisting of the standard reaction mixture, but without the addition of H_2O_2 and $MnSO_4$, was run in parallel.

Glyphosate analysis

A portion (50 μl) of the sample and labelled glyphosate and AMPA internal standards (0.1 μg of each

standard dissolved together in 100 μl of deionised water) were added to an 8-ml glass tube. The sample was completely evaporated under a gentle air stream at 50°C and derivatised by adding TFE (1 ml) and TFAA (2 ml) and holding the sample at 100°C for 1 h (modified from Mogadati et al. 1996). Prior to analysis, the sample was evaporated to dryness in a gentle air-stream and dissolved in ethyl acetate (1 ml). After 1 min of sonication, the sample was analysed by gas chromatography-mass spectrometry (GC-MS).

GC-MS analyses were performed with a Hewlett–Packard 6890 GC system, equipped with a 30 m × 0.25 mm I.D. (0.25 μm film thickness) fused-silica capillary column (HP-5MS 5% phenyl methyl siloxane), a mass spectrometer 5973, a split/splitless injector and software Chemstation, all from Hewlett–Packard (Kista, Sweden). Samples (1 μl) were injected in the splitless mode at 270°C with the oven temperature at 70°C. After 2 min, the oven temperature was raised to 170°C at 30°C min^{-1} and then from 170 to 250°C at 120°C min^{-1}. Helium (N47 grade, 99.997%) was used as the carrier gas at a flow rate of 1.2 ml min^{-1}. The mass spectrometer was operated in the electron impact (EI) mode; the transfer line temperature was 270°C and the manifold temperature 230°C; m/z 126 and 302 were used as the target and qualification ions, respectively, of the AMPA derivative and m/z 411 and 384 were used for the glyphosate derivative. The target and qualification ions for the internal standards were: for AMPA m/z 130 and 306 and for glyphosate 413 and 386. For quantification, the response values for AMPA and glyphosate target ions were calculated in relation to those of the internal standards.

Analysis of the pesticide mixture

The pesticides in Pesticide Mix 34 were analysed at the Department of Aquatic Sciences and Assessment, Section of Organic Environmental Chemistry, Swedish University of Agricultural Sciences.

The samples were filtered through a 0.45 μm filter of PVDF (polyvinylidene fluoride). The filtered sample (5 μl) was injected into a HPLC system (Agilent 1200SL) provided with a SPE-column. The analytes were separated in analytical columns connected in series (Zorbax Eclipse Plus C18, 3.0 × 100 mm, 3.5 μm). The detection was performed in a LC-MS/MS (Agilent G6410A) with electrospray interface

(ES+). The following pesticides and degradation products in the Pesticide Mix 34 could be analysed by this method: atrazine, atrazine-desethyl, chlorotoluron, chloroxuron, crimidine, cyanazine, diuron, fenuron, isoproturon, linuron, metamitron, metazachlor, metobromuron, metolachlor, metoxuron, metribuzin, monolinuron, prometryn, propham, simazine, terbuthylazine and terbutryn.

Results

Degradation of glyphosate by peroxidases and laccases

The effect of laccase on glyphosate degradation is shown in Table 1. In the presence of laccase and ABTS (Lac 2), 40.9% of the glyphosate disappeared after 24 h, whereas 62.8% of the glyphosate was degraded when Mn^{2+} and Tween 80 were added together with the enzyme (Lac 3). A synergistic effect of ABTS, Mn^{2+} and Tween 80 (Lac 4) was observed, where 90.1% of glyphosate disappeared after 24 h. The metabolite AMPA was detected in all the cases where degradation of glyphosate occurred (data not shown). No other metabolites were analysed in the present work, but the equal stoichiometry between AMPA formed and glyphosate degraded suggests that AMPA was not degraded and that there was no or negligible formation of other compounds. No degradation of glyphosate was observed with laccase alone (Lac 1).

Table 2 shows the degradation of glyphosate by MnP. Comparison within the pairs of treatments that differed only in the addition of $MnSO_4$ (treatments M1 vs. M3, M2 vs. M4, M5 vs. M7 and M6 vs. M8 in Table 2) indicates that Mn^{2+} is essential for the oxidative activity of the enzyme, since no significant degradation of glyphosate was detected in the treatments lacking the metal compared with the control. Glyphosate was not degraded when $MnSO_4$ was added without the enzyme (data not shown).

With addition of $MnSO_4$ to the mixture (M3), MnP was able to degrade 53.0% of the glyphosate in the absence of H_2O_2. The degradation increased to 77.6% by addition of H_2O_2 (M4). Total degradation of glyphosate was obtained in the mixtures with MnP, Mn^{2+} and Tween 80 with (M8) or without (M7) H_2O_2. 5 h after the initiation of the reaction, 93%

(M8) and 89% (M7) of the glyphosate had already been transformed, indicating a slight enhancement due to the addition of H_2O_2 ($P < 0.05$) (data not shown). As in the laccase assay, AMPA was stoichiometrically formed in all cases where degradation of glyphosate occurred (data not shown).

Under the conditions tested, neither HRP nor LiP, with or without mediators, was able to transform glyphosate (data not shown).

Degradation of Roundup® Bio by MnP

The effect of MnP on the degradation of glyphosate in its commercial formulated form (Roundup® Bio) was tested using the same reaction mixture as in M7 (Table 2), keeping the same concentration of active ingredient and the same incubation conditions (35°C, 150 rpm). The formulated glyphosate was almost completely degraded after 4 days of incubation (Fig. 1). Simultaneous and stoichiometric formation of AMPA was observed, thus without any indications of it being degraded (data not shown).

Degradation of AMPA by MnP

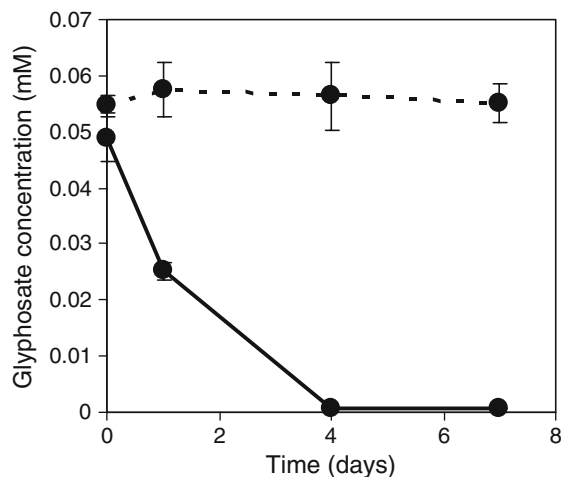The degradation of AMPA by MnP was tested using the same reaction mixture as in M7 (Table 2), under

Fig. 1 Concentration of glyphosate, added as formulated Roundup® Bio, in a reaction mixture with 0.15 U ml^{-1} MnP (*solid line*) and without enzyme (*dashed line*) under in vitro conditions during 7 days incubation at 35°C and 150 rpm. Values are means ± SD ($n = 2$). The reaction mixture contained 0.06 mM glyphosate, 1 mM $MnSO_4$ and 1% Tween 80 in 50 mM Na acetate buffer (pH 4.5)

Fig. 2 Degradation of a mixture of pesticides by MnP under in vitro conditions after 6 days incubation at 35°C and 150 rpm. Values are means ± SD ($n = 3$). The reaction mixture contained 0.45 µg ml^{-1} of each pesticide, 1 mM MnSO$_4$, 1% Tween 80, 0.05 mM H$_2$O$_2$ and MnP (1.5 U ml^{-1}) in 50 mM Na acetate buffer (pH 4.5)
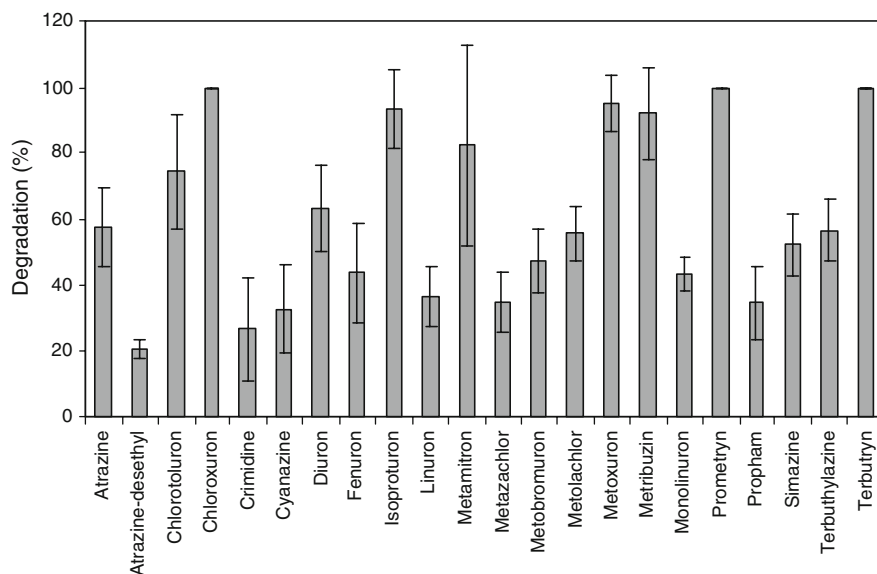
the same incubation conditions but replacing glyphosate by 0.06 mM AMPA. After 5 days of incubation, the concentration of AMPA in the mixture with MnP did not differ from the control without enzyme (data not shown). This confirmed the results above that MnP, under the conditions of our assays, is not able to degrade AMPA.

Effect of MnP on degradation of a mixture of pesticides

All 22 pesticides and pesticide metabolites analysed were degraded to different extents by the enzyme (Fig. 2). Chloroxuron, prometryn and terbutryn were completely degraded, whereas more than 80% of the original amounts of isoproturon, metoxuron, and metribuzin were degraded after 6 days of incubation.

Discussion

The degradation of pesticides and other pollutants by ligninolytic enzymes produced by white rot fungi and by plant peroxidases has been reported and applied in bioremediation and bioprophylactic methods (e.g., Bhunia et al. 2001; Castillo et al. 2008; Mohan et al. 2005; Park et al. 2006). However, the effect of these enzymes on the degradation of glyphosate has so far not been explored. Our results show that whereas HRP and LiP did not have an effect on glyphosate, laccase

and MnP peroxidase were able to rapidly transform the herbicide under in vitro conditions. Glyphosate was not a substrate for laccase since the presence of Tween 80 or ABTS was necessary for the reaction to occur. It is known that such enzyme/mediator systems extend the substrate range of the enzymes. For instance, ABTS mediates the oxidation of non-phenolic compounds of lignin (Bourbonnais and Paice 1990) and HBT the oxidation of phenanthrene (Böhmer et al. 1998) in laccase-catalysed reactions that do not occur in the absence of the mediator.

MnP and laccase initiate peroxidation of unsaturated fatty acids, producing lipid peroxyl or alkoxyl radicals (Böhmer et al. 1998; Hammel et al. 1986; Srebotnik and Boisson 2005). These lipid radicals are highly reactive and can react with other lipid molecules to propagate the oxidation. Tween 80 contains the monounsaturated fatty acid oleic acid. Castillo et al. (2000) found that MnP was able to oxidise the herbicide bentazon when Tween 80 was present, in addition to MnSO$_4$ and H$_2$O$_2$. The degradation of glyphosate by MnP and laccase observed in our assays in the presence of Tween 80 was thus most likely due to a lipid peroxidation reaction.

The presence of MnSO$_4$, or rather probably Mn^{2+}, was a precondition for the degradation of glyphosate by MnP, and even gave an enhanced activity of laccase. The MnP oxidises Mn^{2+} to Mn^{3+}, which in turn oxidises the organic substrate (Kirk and Farrell 1987), in our experiments glyphosate. The enzyme

was able to transform half the herbicide added initially using Mn^{2+} in the mixture, but complete degradation was obtained when Tween 80 was also present in the reaction mixture, even without addition of H_2O_2.

MnP, in combination with $MnSO_4$ and Tween 80, was also able to degrade glyphosate in its formulated form. The other ingredients present in Roundup® Bio (64%) thus did not inhibit the action of MnP, although they seemed to reduce the rate of glyphosate degradation.

Glyphosate is strongly bound to soils (Mamy and Barriuso 2005) and its degradation is considered to be microbial (Pipke and Amrhein 1988) and cometabolic (Torstensson 1985). Microbial degradation occurs via two main pathways. In one, glyphosate is first transformed to sarcosine by a C-P lyase, and then to glycine and formaldehyde by sarcosine oxidase (Kishore and Jacob 1987; Liu et al. 1991; Pipke et al. 1987). In the other, the C–N bond of glyphosate is cleaved by an oxidoreductase, with the formation of AMPA and glyoxylate (Obojska et al. 2002). Subsequently, the enzyme C-P lyase transforms AMPA into inorganic phosphate and methylamine (Cordeiro et al. 1986). Whereas the sarcosine pathway is the most common among isolated strains (Dick and Quinn 1995), this metabolite has not been detected in soils, presumably due to its rapid degradation (Borggaard and Gimsing 2008). Many bacteria transform the herbicide in the absence of a P source (Singh and Walker 2006 and references therein). Some fungi can also use glyphosate as the only P source, e.g., *Penicillium citrinum* (Zboinska et al. 1992), *P. notatum* (Bujacz et al. 1995), *Trichoderma harzianum*, *Scopulariopsis* sp. and *Aspergillus niger* (Krzyśko-Lupicka et al. 1997) and as the only N source, e.g., *P. chrysogenum* (Klimek et al. 2001).

Glyphosate degradation by enzymes from white rot fungi has not previously been reported. Our present work shows that two of the ligninolytic enzymes produced by these microorganisms, MnP and laccase, are very efficient in the transformation of glyphosate, and lead to AMPA formation and accumulation. For MnP, none of the mediators tested in our experiment (Mn^{2+}, Tween 80 and H_2O_2), was suitable for AMPA degradation. Further studies are thus needed to explore whether such mediators can be found.

AMPA is frequently found in soils that have been exposed to glyphosate, and its degradation seems to be slower than that of the parent compound (Torstensson 1985). The only enzyme responsible for the transformation of glyphosate to AMPA that has been identified so far is the glyphosate oxidoreductase from *Ochrobactrum anthropi*, which is the basis of the glyphosate-tolerant Roundup Ready crops (Barry et al. 1992). Our results show that the transformation of glyphosate to AMPA in soil can also be explained by the activity of enzymes such as MnP and laccase from white rot fungi. In addition, several actinomycetes, notably streptomycetes, are able to produce extracellular peroxidases involved in degradation of lignocelluloses (Arias et al. 2003; Trigo and Ball 1994; Tuncer et al. 2004). As common soil-dwelling microorganisms (Goodfellow and Williams 1983), their contribution to the degradation of glyphosate may thus also be important.

Due to their non-specificity, the degradation potential of ligninolytic enzymes is not restricted to glyphosate among the pesticides. The herbicides bentazon and isoproturon can be degraded individually by MnP (Castillo et al. 2000, 2001) and both isoproturon and metribuzin, added in a mixture with methabenzthiazuron, have been shown to be dissipated in straw cultures of *Phanerochaete chrysosporium* (Castillo and Torstensson 2007). Interestingly, in the present work, the simultaneous degradation of 22 pesticides in a mixture was obtained by the action of MnP in the presence of Tween 80 and $MnSO_4$, with degradation values varying from 20 to 100%.

In conclusion, purified MnP and laccase were able to degrade the herbicide glyphosate to AMPA under in vitro conditions. In addition, MnP degraded the herbicide in a commercial formulation (Roundup® Bio) in the presence of $MnSO_4$ and Tween 80. Under the same conditions, MnP was also able to simultaneously degrade 22 pesticides and degradation products in a mixture. However, no transformation of AMPA by MnP was observed under the conditions tested.

Our results highlight the potential of ligninolytic enzymes to degrade pesticides and may contribute to better understanding of the often largely different persistence times obtained for pesticides in different soils.

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