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Changes in bacterial communities by post-emergent herbicides in an Andisol fertilized with urea as revealed by DGGE

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ABSTRACT

Large amounts of urea and various herbicides are applied to Chilean Andisols by farmers; however, how different herbicide types affect bacterial communities in urea-fertilized soils remains unknown. In this study, the effect of post-emergent herbicides (MCPA, flumetsulam, fluroxypyr, triclopyr, clopyralid and picloram) on total bacterial (TB) and ammonia-oxidizing bacteria (AOB) communities was investigated in a urea-fertilized soil microcosm using denaturing gradient gel electrophoresis at days 1 and 15 of incubation. The residual concentrations of herbicides in microcosm soils were measured by high-performance liquid chromatography. Differences in bacterial communities were analyzed and visualized with non-metric multidimensional scaling. All of the tested herbicides decreased in microcosm soils, reaching 46–98% of dissipation at 15 days. Changes in the TB and AOB communities were variable, depending on the urea and herbicide dosage, herbicide type and sampling time. Under the recommended dose of urea (570 mg kg^{-1} soil), soils that were treated with MCPA and triclopyr showed significant changes in TB and AOB community structures compared to control soils at both sampling times. When a 2-fold-higher recommended dose of urea (1140 mg kg^{-1} soil) was applied, the TB communities were mainly affected by flumetsulam and picloram at both sampling times. Changes in the AOB communities were mainly observed in soils that were treated with MCPA and fluroxypyr under both urea doses. This study determined which herbicides (and doses) significantly affect the bacterial communities in urea-fertilized soils. This evidence can be very useful when management practices are being designed, revised and/or established in Andisols.

1. Introduction

In modern agriculture, large amounts (3.75 million tons in 2000 and an estimated 6.55 million tons for 2020) of pesticides are commonly applied to soils to control weeds and to improve the quality of crops and pasture systems (Tilman et al., 2001). However, the extensive and long-term application of a wide variety of herbicides is of increasing concern because of their environmental impact and effect on soil microorganisms, which are essential for soil fertility and processes in terrestrial ecosystems, such as the nitrogen (N) and phosphorus (P) cycles. Some herbicides are accumulated in the top soil layer, where most of the microbiological activities occur, affecting the biomass, diversity, respiration, and enzymatic activity of soil microorganisms, as well as biogeochemical processes, such as ammonification, nitrification and denitrification (Cycoń and Piotrowska-Seget, 2007; Hernández et al., 2011; Jezierska-Tys and Rutkowska, 2015; Kara et al., 2004; Paulin et al., 2011).

The post-emergent herbicides MCPA, flumetsulam, picloram, clopyralid, fluroxypyr, and triclopyr are commonly applied on permanent pastures to control broadleaf weeds in Chile. MCPA is chemically classified as a phenoxyacetic herbicide, which has a carboxylic ionizable group. Flumetsulam is classified as a sulfonanilide herbicide, whereas clopyralid, fluroxypyr triclopyr, picloram are classified as pyridine herbicides. All of mentioned herbicides have a carboxylic acid group, except flumetsulam which has an acidic H atom, bound to N atom. They are also considered as acidic herbicides with low pK_a values (Table 1) and exist mainly as anions in the soil environment. In this context, these herbicides are weakly sorbed to soil through hydrophobic interactions and hydrogen bonding with soil organic matter (OM). Hydrophilic

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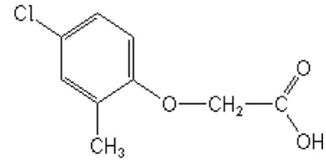
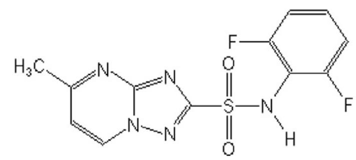
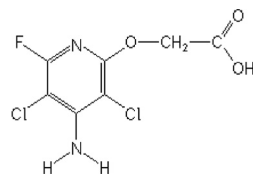
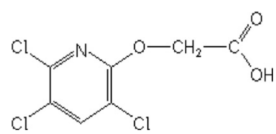
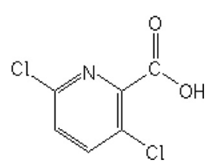
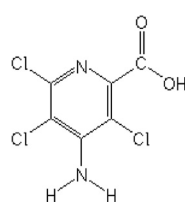
Table 1
Some properties of post-emergent herbicides used in this study.

Herbicide	Formula	Purity (%)	Sw[a] ($g L^{-1}$)	Log P[b]	pK_a
MCPA (2-methyl-4-chlorophenoxyacetic acid)		99.0	734	−0.81	3.7
Flumetsulam (*N*-(2,6-difluorophenyl)-5-methyl[1,2,4]triazolo[1,5-a]pyrimidine-2-sulfonamide)		98.8	49	−0.68	4.6
Fluroxypyr ([(4-amino-3,5-dichloro-6-fluoro-2-pyridinyl) oxy]aceticacid)		99.5	0.09	0.04	2.9
Triclopyr ([(3,5,6-trichloro-2-pyridinyl) oxy]aceticacid)		99.5	8.0	4.62	4.0
Clopyralid (3,6-dichloro-2-pyridinecarboxylic acid)		98.8	143	−2.63	2.0
Picloram (4-amino-3,5,6-trichloro-2-pyridinecarboxylic acid)		98.8	0.6	−1.92	2.3

[a] Water solubility.
[b] Log P, octanol-water partition coefficient.Source: Tomlin (1995) and PPDB (The Pesticide Properties DataBase), 2013.

mechanisms have been proposed for explaining sorption of acidic herbicides in soil of variable-charge, through anion exchange and Ca^{2+}-bridging processes on oxide surfaces (Hyun and Lee, 2004; Kah and Brown, 2006).

Andisols are the predominant soil type supporting agricultural and livestock production in southern Chile and widely distributed throughout the world. Andisols are ash-derived volcanic soils, which consist of minerals with a low degree of crystallization and a variable pH-dependent charge. The mineral fraction presents high content of allophone, imogolite and ferrihydrite, and in the most superficial horizons Al-humus and Fe-humus complexes. Andisols are characterized by high OM content, low pH, a high specific surface area, a low bulk density and high water retention capacity (Escudey et al., 2004). The behavior of post-emergent herbicides in Andisol is also influenced by their chemical structure and soil properties (Palma et al., 2015a,b).

Another extensive practice in modern agriculture is the application of nitrogen-based fertilizers, which strongly affect the composition, biomass and activity of soil microorganisms (Berthrong et al., 2014; Geisseler and Scow, 2014). In this context, N fertilization *per se* or combined with other practices (mowing, organic amendments, etc.) induces changes in the bacterial community composition in soils, including soil bacteria that are involved in N cycling, such as ammonia-oxidizing microorganisms (Chen et al., 2014; Hallin et al., 2009; Shen et al., 2010; Webster et al., 2002). In relation to interactions between N fertilization and herbicide efficacy, Sønderskov et al. (2012) reported that the N rate (20, 80 and 120 $kg\,N\,ha^{-1}$) affects the herbicide efficacy of some but