3 B.5 - Glyphosate resistant weeds in Europe: a review

A. Collavo[1], C. Gauvrit[2], N. Muelleder[3], M. Sattin[1] & R. De Prado[4]
[1] Consiglio Nazionale delle Ricerche, Istituto di Biologia Agro-ambientale e Forestale, Viale dell'Università 16 - 35020 Legnaro (PD) Italy. E-mail: alberto.collavo@ibaf.cnr.it
[2] INRA, UMR 1210 Biologie et Gestion des Adventices, F-21000 Dijon, France.
[3] Monsanto International SARL
[4] Cordoba University, Dep. Quimica Agricola y Edafologia, Cordoba, Spain.

Abstract: Glyphosate is the world's most widely used herbicide, with many registrations in agricultural, urban and semi-natural environments. Glyphosate is an important tool especially for broad-spectrum weed control and inter-row vegetation management in perennial crops like olive and citrus groves, orchards and vineyards. Despite the frequent use in these crops, there are only a few confirmed cases of glyphosate resistant weed populations in Europe. The first reported European case dates back to 2004 and involved four populations of *Conyza bonariensis* found in southern Spanish olive groves. The glyphosate rates required to control resistant populations were 7 to 10 times higher than those needed to control the susceptible populations. In 2006 a resistant population of *C. canadensis* was reported in southern Spain while another one was claimed in the Czech Republic in 2007. The resistance in Czech *Conyza* is currently still under investigation. Recently glyphosate-resistant *Lolium rigidum* was found in French vineyards (2005 and 2007) and in Spanish citrus groves (2006), while resistant *L. multiflorum* was identified in Spanish olive groves (2006). New cases of resistant *Lolium* spp. are reported from Italian vineyards and olive groves. In all these cases *Lolium* plants were not controlled at recommended glyphosate field rates. Although glyphosate resistant weed populations in Europe are only a few, the sole reliance on glyphosate for weed control especially in perennial crops bears the risk of selecting more resistant populations. Resistance management should be based on principles of Good Agricultural Practices and Integrated Weed Management:
- use the right rate at the right time;
- apply glyphosate when the plants are more susceptible and the environmental conditions are favourable for its uptake;
- integrate herbicides with different mode of action (e.g. a selective or residual herbicide) and mechanical weed control in the weed control program.
The new EU legislation on plant protection products is likely to decrease the number of active ingredients and herbicidal mode of actions available for weed control and therefore introduces new challenges for managing weed resistance.
Keywords: *Conyza* spp., *Lolium* spp., selection, sustainability, evolution, IPM, weed, herbicide.

INTRODUCTION

Glyphosate was first marketed in 1974 and gradually has become the most widely used herbicide in the world. It is used in a wide range of situations, controlling efficiently a large range of weed species, it has a benign environmental profile and offers good value for price. It is widely adopted in annual and perennial crops, in till and no-till systems, in pre-plant applications and, where available, in glyphosate resistant crops (GRCs). It is also an important tool to manage weed biotypes resistant to other modes of actions (e.g. ALS or ACCase inhibitors).

The target site of glyphosate is the 5-enolpyruvylshikimate-3-phosphate (EPSP) synthase, which catalyses the aromatic amino acid biosynthesis. Glyphosate has always been considered a low-risk herbicide for selecting resistance (BECKIE, 2006), at least until the wide spread adoption of GRCs in north and south America. In fact the first case of glyphosate resistance was reported in 1995, more than 20 years after its introduction. Since 1996, glyphosate resistance has been reported in 15 weed species worldwide, however just four species are involved in Europe: *Conyza bonariensis* L., *C. canadensis* L., *Lolium multiflorum* Lam. and *L. rigidum* Gaud. (HEAP, 2009). The reasons for very moderate occurrence of resistant populations is probably due to the fact that the selection is exerted just on the fraction of emerged plants from the soil seed bank, thus reducing the selection pressure on the overall population. Further on there is no residual activity of glyphosate, so the exposure of weeds to the active is not long lasting.

In situation having just one treatment per year and/or controlling weeds using different strategies results in the maintenance of reasonable level of diversity which, in turn, positively impacts glyphosate sustainability (POWLES, 2008).

In Europe significant glyphosate selection pressure is present in perennial crops (e.g. vineyards, orchards, olive and citrus groves) where glyphosate is often exclusively used two or three times per year. Above that the comparable low price reduces the interest on other chemicals. This repeated and exclusive use of glyphosate in perennial crops is the reason for the development of resistant biotypes as it happen with other situations of high selection pressure for resistance.

STATUS OF GLYPHOSATE RESISTANCE IN EUROPE

The first case of glyphosate resistance reported in Europe dates back to 2004 involving four *Conyza bonariensis* populations found in olive groves located in Andalusia (southern Spain) in the provinces of Seville, Huelva and Cordoba (URBANO et al., 2007). The resistant populations had been repeatedly treated with glyphosate (at least one application per year for a period of 4–6 years) and the orchard was characterised by no-tillage and drip irrigation. The resistance index ranged between 3.5 and 10.5. Two different resistance mechanisms appeared to be involved. The first was an impaired herbicide translocation while the second mechanism was a higher basal mRNA level of the glyphosate target site enzyme which lead to an increased EPSP synthase activity (DINELLI et al., 2008). Two out of the four populations tested had both mechanisms operating. Two more cases of resistant *C. bonariensis* were reported later.

In an orchard in Spain there was also a case of *C. canadensis* reported in 2006. In total, Spanish glyphosate resistant *Conyza* spp. are believed to be spread on 3000-5000 ha (URBANO, personal communication).

A different case of increased *C. canadensis* susceptibility was reported for a Czech weed population collected in 2007 along a railway in Prague-Bubny and Prague-Libeň areas. The population had been continuously treated with glyphosate (720 g a.e. ha^{-1}) since 2000. An experiment in growth room outlined that a dose of 1800 g a.e. ha^{-1} completely controlled this population. Further investigations are needed to precisely determine the level of increased tolerance/resistance.

The other main weed involved in European glyphosate resistance is *Lolium*. *L. multiflorum* glyphosate resistant biotypes were collected in Spanish olive groves in 2006 while *L. rigidum* resistant populations were found after a survey all over Spain. A population from Castilla/Leon had a resistant index of 5.3 and presented multiple resistance to fenoxaprop and imazamethabenz and the resistance

mechanism was attributed to a slower rate of absorption and translocation (CALHA et al., 2008). All the above *Lolium* populations survived a dose of 1440 g a.e. ha^{-1} (DE PRADO, unpublished data).
Resistant *L. rigidum* has also been confirmed in France, the first report dates from 2005 and in 2007 two more cases were added. All these populations have been selected in vineyards.
In Italy *Lolium* is present from north to south, the populations from north can be attributed to the species *L. multiflorum* while those from south to *L. rigidum*. Intermediate forms are widespread because of the high hybridisation typical for this genus. Two populations of *Lolium* spp. resistant to glyphosate have been found, one in northern vineyards and the other in southern olive groves. First molecular investigations indicate that resistance mechanism involved in the southern populations is related to an altered target site. Survival of plants from this population treated with a dose of 1440 g a.e. ha^{-1} ranged from 56 % to 88 %. Glyphosate resistance through pollen flow may occur in those species characterised by obliged out-crossing (like *Lolium*). In northern Italy the presence of resistant plants of *Lolium* have been observed (COLLAVO et al., unpublished data) in a population which has always been controlled mechanically. These biotypes are growing near a site with glyphosate resistant *Lolium* biotypes.
A list of current glyphosate resistant weed biotypes in Europe is shown in Table 1.

Table 1. Glyphosate resistant weeds in Europe.

Country	Species	No. biotypes	First year report
Spain	*Conyza bonariensis*	6	2004
	Conyza canadensis	1	2006
	Lolium rigidum	1	2006
	Lolium multiflorum	1	2006
Czech Republic	*Conyza Canadensis*(*)	1	2007
France	*Lolium rigidum*	3	2005
Italy	*Lolium* spp.	2	2008

(*) to be confirmed

CONCLUSIONS

Glyphosate is the most widely used non-selective herbicide. It is a very important tool in numerous cropping systems, both in arable and in perennial crops and due to its broad-spectrum efficacy and ecotoxicologial properties it is frequently used. Four species and four countries are affected by glyphosate resistant weed biotypes in Europe.
To maintain its efficacy glyphosate should be used carefully and needs to be integrated to a range of weed control methods. Increasing weed control diversity is the best strategy to preserve any chemical from selecting resistant plants (SATTIN, 2005).
The way glyphosate is used determines the risk for selecting resistance. A solution to avoid, or at least delay, glyphosate resistance could be to limit the number of applications to one per year (BECKIE, 2006). But in order to avoid resistance development and to control resistant weeds, the principles of Good Agricultural Practices and Integrated Weed Management are most probably more successful ways to address the problem: by applying glyphosate when the plants are more susceptible; by using cultural practices like tillage and/or mowing in complement to chemical control; by using herbicide sequences and tank mixtures with different modes of action and by absolutely avoiding to spread resistant pollen and/or seeds.

It is not surprising that the populations resistant to glyphosate in Europe belong to the most resistant-prone genera: *Conyza* and *Lolium*. These genera are characterised by high seeds production and cross-pollination.

Depending on the regulatory approval status in the respective countries resistant *Conyza* can be controlled by tank mixing glyphosate with active ingredients like fluroxypyr, flazasulfuron, aminotriazol or MCPA. For *Lolium* aminotriazol, cycloxydim, clethodim and flazasulfuron are options.

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