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Research Article

Herbicides Use in Wheat Crop in Tunisia: Trends, Variability and Relation with Weed Resistance Development

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Abstract

In this study, we investigated the situation of herbicide use in wheat in Northern Tunisia and its relation with the development of resistant weed populations. It is based on a synthesis of available data concerning Tunisian imported herbicide quantities, treated wheat areas and herbicides used by farms surveyed in Northern Tunisia and their correlation with rainfall and yields. Herbicide-treated areas were classified into two categories: the areas treated with the 2,4-D herbicides and the areas treated with graminicides and large spectrum herbicides. The first herbicides used in the 70s are the anti-broadleaf herbicides (2,4-D). Since the 80s, graminicides and large spectrum herbicides began to be more and more used for grass weed control. In 2008/2009, graminicide-treated areas represent above twice those treated with 2,4D. However, year-to-year fluctuations are observed and seem to be correlated to autumn rainfall.However, the variety of commercial herbicides corresponds to a small number of molecules and particularly modes of action. A survey conducted in northern Tunisia showed that the majority of herbicides used by farmers for weed control in wheat belong to ALS and ACCase inhibiting herbicides. Consequently, problems of resistant weed populations (e.g. rye grass) have already been observed and intensive use of the molecules used today can only be a short-term solution. A rational management of these molecules is essential to maintain their effectiveness as long as possible.

Keywords: Graminicides, 2,4D, yield, rainfall.

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Introduction

In Tunisia, cereals represent 1.5 million hectares, of which more than 70% is wheat. They correspond to 35% of arable land. Wheat production is a priority in Tunisia and its increase is politically encouraged to reach national self-sufficiency. Weeds represent a continuing problem in Tunisian cereal production and are one of the limiting growing factors (Gressel et al 2004; Latiri et al 2010). Therefore, weed control is an essential component of productive agriculture.

Herbicides are the major tool of controlling weed growth. Easy to apply and rather inexpensive, compared to other weed control methods such as hand weeding, chemical control products have proved to be extremely efficient and reliable in a very large number of cases, on large surface areas (Powles and Shaner 2001). Consequently, agriculture has come to rely on herbicides as the major instrument of controlling undesired weed growth and it became one of the inputs used in the green revolution in developing countries (Briggs et al 2009). However, today, with the increasing awareness of their negative impacts and the demonstration of undesirable adverse effects on ecosystems, the systematic use of herbicides is being called into question (Zoschke and Quadranti 2002; Mamy et al 2005). In fact, the intensive use of herbicides in agriculture represents a significant risk to the environment, mainly to the soil and water qualities, and human health (Weisenburger 1993; Guzzella et al 1996). Furthermore, it has resulted in the selection of herbicide resistant weed populations which can severely hamper herbicide-based weed control, thus leading to the need for a more accurate use of these chemical products (Powles et al 1996; Heap 2011). Throughout the world, over 194 weed species have thus far evolved resistance to at least one herbicide (Heap 2011). In Tunisia, the first report of evolved herbicide resistance concerned *Lolium rigidum* (ryegrass) populations collected in wheat fields in Northern Tunisia in 1996 and found to be resistant to Acetyl-Coenzyme A carboxylase-inhibiting (ACCase) herbicides (Souissi et al 2004).

To better understand the risk of development of resistance that herbicide use can generate, this work aims at presenting a synthesis of available data on herbicides use in wheat fields in Northern Tunisia. This region is the largest and most stable wheat production area in Tunisia, being about 60% of the total area harvested and about 68% of cereal production. Moreover, Northern Tunisia is the region where cereal production is most intensified and where most of the problems of the effectiveness of herbicides have been reported (Souissi et al 2004). Herein, we analyzed the evolution of wheat herbicide-treated areas and herbicides quantities used first at the whole country level, then for the Northern part of the country, with regional statistical data and surveys of farmers conducted in Northern Tunisia. Relationships with wheat yield and with rainfall are considered as well as molecules and association used by farmers.

Materials and methods

Data sources and analysis

For the analysis of the herbicides usage in Northern Tunisia, yearly data concerning the herbicide-treated areas, wheat yields and rainfall were collected from the Ministry of Agriculture and Environment from 1974/1975 to 2008/2009 in the governorates of Béja, Jendouba, El Kef, Siliana, Bizerte, Tunis and Zaghouan (Figure 1), which are characterized to be a sub-humid to semi-arid climate.

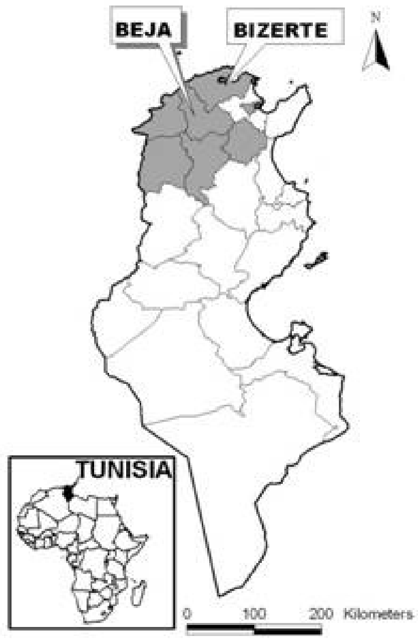

Figure 1- Map of Tunisia showing in grey Northern governorates and indicate the two governorates where survey was conducted (Bizerte and Béja)

In the absence of data concerning herbicides usage at farm level, data concerning the amounts of herbicides imported in Tunisia were collected from the National Statistics Institute from 1991 to 2008.

In Tunisia, wheat-growing season is extending from November until May. For each governorate, the amount of rainfall of each wheat-cropping season was calculated based on the monthly rainfall available for different meteorological stations. Wheat yields were estimated through a weighted average of bread and durum wheat and their respective areas.

Farmer's survey

To better understand cultural practices and herbicide applications of farmers, a survey of fifty farmers was conducted in 2007 in the governorates of Bizerte and Béja. Surveys were conducted randomly and were carried out to cover the entire areas of the two governorates surveyed and to determine if problems related to the effectiveness of herbicides could be observed.

The frequency of use of each herbicide was expressed as the number of applications of each herbicide by the total of applications, thus reflecting the most used herbicides by farmers.

Results and discussion

Evolution of herbicides usage in Tunisia since 1991

Presently, close than 1000 pesticides corresponding to 398 active ingredients are marketed in Tunisia. Among these pesticides, herbicides represent 26% of the total active ingredients, with 177 products and 103 active ingredients (PAN 2006).

The large majorities of pesticides are imported as active ingredients or as formulations, and are mostly used in the agriculture sector. Pesticide importations reach 3600 tons year^{-1} (PAN 2006). Herbicides occupy the third place (23%) after insecticides (43%) and fungicides (31%).

Given the nature of available data, the evolution of herbicides usage was estimated throughout the evolution of active ingredient import. Herbicides usage has significantly increased since 1990. In 18 years, the amount of herbicide has increased from 422 tons in 1990 to 1265 tons in 2008 (Figure 2). From 1990 to 1998, imported herbicides are mainly sulphuric acid and germination inhibitors. Since 1999, another group of compounds, the triazines, the dinitroanilines, ureas, phenoxy acids and acetolactate synthase inhibitors are added to the list of imported herbicides.

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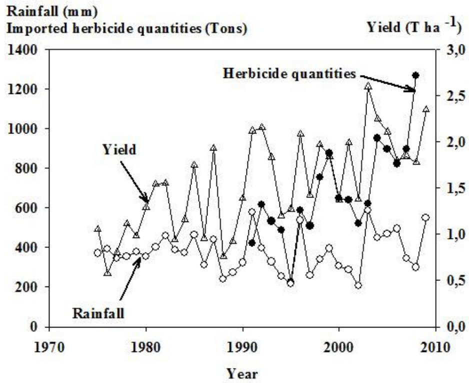


Figure 2- Evolution over years of rainfall, wheat yields and imported herbicide quantities in Northern Tunisia

Despite this increase, some years have seen considerable reductions in imports. The reductions observed during the 1994/1995 and 2001/2002 agricultural campaigns are corresponding to severe drought years in Tunisia (Figure 2). When the expected yield is low, herbicides are rarely or in low dosages used by farmers in order to minimize production costs. Indeed, a significant correlation between the amount of herbicides imported and wheat yields in the Northern region was noted ($r = 0.52$, $P < 0.05$, $n = 18$).

Statistical data collected represent present the quantity of imported herbicides per major families of herbicides. They do not therefore allow for monitoring the evolution of the use of certain active ingredients. In addition, the indicator "tonnage" used in national statistics does not reflect the evolution of active substances. In fact, modern herbicides are used with a low rate (few grams per hectare for the acetolactate synthase inhibiting herbicides). In order to get a better and clearer idea of herbicide usage, we analyzed the evolution of herbicide treated areas in Northern Tunisia over time.

Evolution of herbicide-treated wheat areas in Northern Tunisia since 1975

Herbicide-treated areas are classified into two categories: (i) the areas treated with the 2,4-D herbicides and (ii) the areas treated with graminicides and large spectrum herbicides.

During the 1974/1975 growing season, surfaces treated with 2,4-D were nearly 111 500 ha against only almost 3500 ha for graminicide-treated areas, which represent respectively 17.3% and 0.5% of wheat areas in Northern Tunisia (Figure 3). Since 1975, it can be observed that 2,4-D-treated areas have fluctuated in size, with an average of 135000 ± 53 000 ha, while the graminicide-treated areas have continually increased (Figure 3).

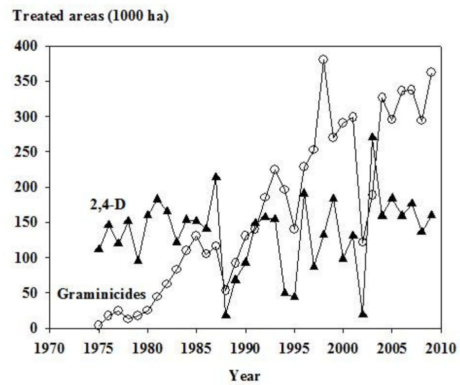
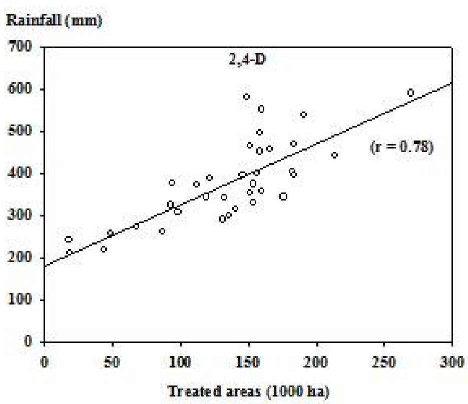


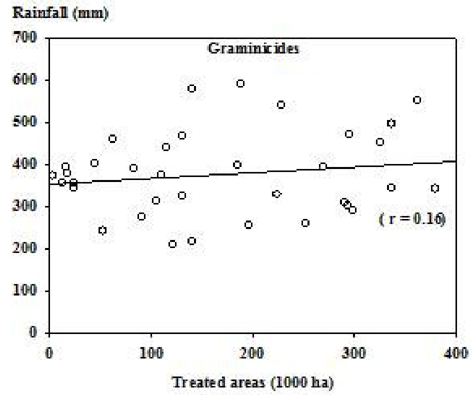
Figure 3- Evolution over the time of 2,4-D-treated areas and graminicide-treated areas in Northern

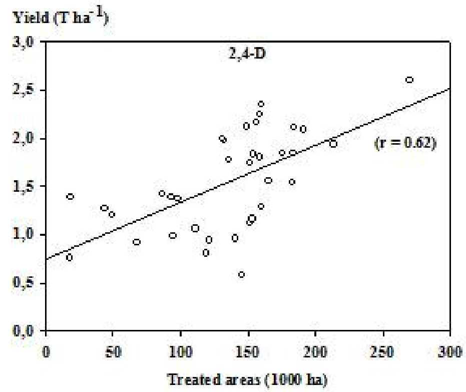
The graminicide treated areas have increased to 362450 ha in 2008/2009 and are slightly above twice those treated with 2,4-D (160 000 ha in 2008/2009; Figure 3). Selective graminicides seem to be intensively used since the mid of 1980's (110 000 ha in 1983-1984). Globally, the part of herbicide-treated wheat areas increase from 27% of the sown wheat areas in Northern Tunisia in 1978 to 85% in 2009.

The wheat intensification since the early 1960's in Tunisia has seen the arrival of graminicide selective herbicides such as sulfonylureas and substituted ureas. Until then, the only herbicides used had been synthetic phytohormones such as 2,4-D, which controlled many types of broadleaf weeds but were ineffective on grasses and allowed the extension of grass weeds, which became the most important class of weeds (Guillou 1975; Rondia et al 1976). In the 1980's, chemical control demonstrations of grass weeds in wheat with these selective products were introduced in Northern Tunisia in nearly 1200 farms (Carème et al 1990).

However, the use of herbicides varies from year to year and fluctuations in the herbicide-treated areas could be observed (Figure 3). In Tunisia, 93% of cereal crops are conducted in rainfed conditions and variations in the treated areas could be attributed to rainfall (Latiri et al 2010). In the case of the herbicide 2,4-D, a highly significant correlation was observed between herbicide-treated areas and the average rainfall between November and May (r = 0.78, P <0.01, n = 35) (Figure 4), period that coincides with wheat growing season and vegetative growth. This moist period is favorable also to weeds growing which correspond to optimal efficacy of 2,4-D. Consequently, if rainfall is low, the potential yield is reduced; thus, rainfall determines the farmer's decisions regarding weed control. This might also explain the highly significant correlation between the areas treated with the 2,4-D and wheat yields (r = 0.62, P <0.01, n = 35) (Figure 4), both of them being linked to rainfall and weed control allowing a better yield.







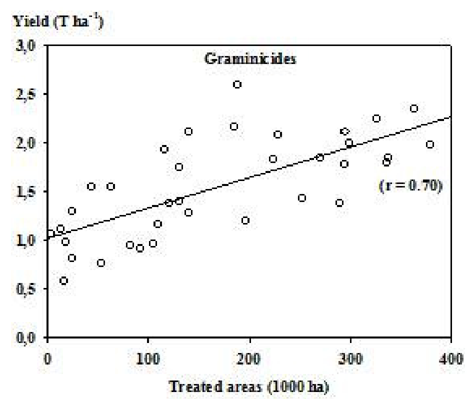


Figure 4- Relationship between herbicides treated areas and rainfall (up) and wheat yield (down)

In the case of the graminicides, a significant correlation between the herbicide-treated areas and yields was observed (r = 0.70, P <0.01, n = 35), but not in relation to the average rainfall (r = 0.16, NS, n = 35)

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(Figure 4). Nevertheless, significant reductions of the graminicide-treated areas were recorded during the years of severe drought (i.e. during 1987/1988, 1994/1995 and 2001/2002; Figure 3).

Hence, farmers' decisions for use of herbicides are different for 2,4-D and for graminicides. 2,4-D use is related to rainfall and to farmers looking for increased yield while graminicides are starting to be used in a more systematic way, unless there is a drought risk. In the past three decades, graminicide treatments were almost not used and farmers were trying to preserve some of the grass to be grazed by sheep in semi-arid areas.

Marketed cereal herbicides in Tunisia

The list of herbicides used in cereal crops in Tunisia has been compiled from the "Phytosanitary Guide of Tunisia (2009)" (ATPP 2009), which lists the authorized and marketed pesticides in Tunisia. The data have been reclassified according to their efficacy (broadleaf weeds, grass weeds, and large spectrum herbicides) and grouped by active ingredient.

Among the 177 registered herbicides in Tunisia, 43 products corresponding to 26 active and 11 chemical families are used in wheat. In the case of the broadleaf, grass and the large spectrum herbicides, there are 18, 10 and 4 formulations belonging to 4, 1 and 4 modes of action respectively, either alone or in combination (Table 1).

In the domain of herbicides, any genuinely mode of action in the world was not discovered for several years. It is unlikely that any new herbicides with new modes of action will be marketed in the near future (Moss 2006). The new active substances belong to families that are already known. In Tunisia, the comparison between the list of herbicides in wheat in 2009 (ATPP 2009) with that of 2006 (ATPP 2006), reveals that two new graminicides molecules have been approved: "prosulfocarb" from the family of thiocarbamates and "pinoxaden" from the phenylpyrazolines family. These two molecules belong to groups which modes of action are already known (Table 1). This molecule is already marketed in Australia, UK, the USA and Europe (Moss 2006; Yong et al 2007; Yu et al 2007).

Table 1: Herbicides used in cereal crops in Tunisia (ATPP, 2009)

Weed	Active substance	Family	Mode of action	Example trade name
Dicotyledonous	2,4-D + Carfentrazone-ethyl	Phenoxy-carboxylic-acids + Triazolinones	Synthetic auxins +PPO inhibitors	Aurora plus
	2,4-D + Florasulam	Phenoxy-carboxylic-acids + Triazolopyrimidines	Synthetic auxins +ALS inhibitors	Mustang 306
	2,4-D acid + Metosulam	Phenoxy-carboxylic-acids + Triazolopyrimidines	Synthetic auxins +PPO inhibitors	Factor
	2,4-D sel d'amine	Phenoxy-carboxylic-acids	Synthetic auxins	Sanhormone
	2,4-D - Ester butylglycol	Phenoxy-carboxylic-acids	Synthetic auxins	Desormone lourd
	Florasulam + Flumetsulam	Triazolopyrimidines	PPO inhibitors	Derby
	Flucarbazone-Sodium	Triazolinones	PPO inhibitors	Everest 70 WG
	Mecoprop - P	Phenoxy-carboxylic-acids	Synthetic auxins	Gallium A
	Amidosulfuron + Iodosulfuron	Sulfonylureas	ALS inhibitors	Sekator
	Tribenuron methyle	Sulfonylureas	ALS inhibitors	Oscar
	Thifensulfuron methyl + Tribenuron methyl	Sulfonylureas	ALS inhibitors	Harmony extra
	Triasulfuron + Dicamba	Sulfonylureas + benzoic acids	ALS inhibitors + synthetic auxins	Zoom
	Bentazone	Benzothiadiazines	Photosynthesis inhibitors	Basagran
	Dicamba - D	Benzoic acids	Synthetic auxins	Banvel 4 S
	Dicamba + 2,4-D	Benzoic acids + Phenoxy-carboxylic-acids	Synthetic auxins	Dialène Super
	Terbutryne + Triasulfuron	Triazines + Sulfonylureas	Photosynthesis inhibitors + ALS inhibitors	Logran extra
	Methabenzthiazuron	Substituted ureas	Photosynthesis inhibitors	Tribunil
	Metoxuron	Substituted ureas	Photosynthesis inhibitors	Dosanex insistent
Monocotyledonous	Diclofop-methyl + fenoxaprop-P-ethyl	Aryloxyphenoxypropionates	ACCase inhibitors	Illoxan Super New
	Clodinafop-propargyl + cloquintocet-2-Mexyl	Aryloxyphenoxypropionates	ACCase inhibitors	Topik 100
	Tralkoxydim	Cyclohexanediones	ACCase inhibitors	Grasp
	Diclofop-Methyl+ Bromoxynil	Aryloxyphenoxypropionates + Hydroxybenzonitriles	ACCase inhibitors	Illoxan B
	Diclofop-Methyl+ fenoxaprop-P-ethyl	Aryloxyphenoxypropionates	ACCase inhibitors	Illoxan Super
	Fenoxaprop-p-Ethyl	Aryloxyphenoxypropionates	ACCase inhibitors	Puma Super
	Pinoxaden	Phenylpyrazolines	ACCase inhibitors	Axial 045
	Pinoxaden + Clodinafop-propargyl	Phenylpyrazolines + Aryloxyphenoxypropionates	ACCase inhibitors	Traxos 45 EC
	Diclofop-Methyl	Aryloxyphenoxypropionates	ACCase inhibitors	Illoxan 36
	Prosulfocarb	Thiocarbamates	Lipid synthesis inhibitors - not ACCase inhibitors	Boxer
Dicotyledonous and monocot	Fenoxaprop-P-ethyl + Iodosulfuron Methyl-sodium	Aryloxyphenoxypropionates + Sulfonylureas	ALS inhibitors + ACCase inhibitors[a]	Puma Komplete
	Iodosulfuron + Mesosulfuron	Sulfonylureas	ALS inhibitors	Atlantis
	Mesosulfuron-Methyl + Iodosulfuron	Sulfonylureas	ALS inhibitors	Amilcar
	Chlortoluron	Substituted ureas	Photosynthesis inhibitors	Tolurex 500

[a] PPO : Protoporphyrinogen oxidase, ALS : acetolactate synthase, ACCase : acetyl coenzyme A carboxylas

Herbicides used by surveyed farmers

Broadly speaking, the list of herbicides does not really reflect herbicides used by farmers. In France, for example, only 42% of registered pesticides are sold (Aubertot et al 2005).

In the absence of statistics concerning the amounts of pesticides sold that would indicate among the herbicides marketed those most used by farmers, which illustrate the broad trends of the market and put them in relation with some events such as the development of resistance, a survey based on their herbicide program was directly conducted in the regions of Bizerte and Béja. Among the 25 anti-broadleaf, 12 graminicides and 6 large spectrum products approved in Tunisia, only 3, 3 and 2 products, respectively, are used by the farmers interviewed (Table 2).

The anti-broadleaf herbicides and graminicides represent 22.3% and 20.3% of treatments carried out by the interviewed farmers, respectively. The two herbicides: Puma Komplete (Fenoxaprop-P-ethyl, Iodosulfuron Methyl-sodium) and Amilcar (Mesosulfuron-Methyl, Iodosulfuron) seem to be the most used by farmers. Together, they account for 57.4% of the number of herbicides applications.

Table 2: Frequency of the broad leaf, graminicides and large spectrum herbicides used by farmers

Group	Trade Name	Active substance	Treatment frequency
broad leaf	Mustang	2,4-D + Florasulam	13.1
	Oscar, Grandstar	Tribenuron methyl	9.2
Graminicides	Puma super	Fenoxaprop-p-Ethyl	7.3
	Topik 100	Clodinafop-propargyl	7.3
	Illoxan super	Diclofop-methyl + Fenoxaprop-P-ethyl	5.7
Large spectrum	Amilcar	Mesosulfuron-Methyl + Iodosulfuron	18.5
	Puma Komplete	Fenoxaprop-P-ethyl + Iodosulfuron Methyl-sodium	38.9

Herbicides use and resistance development

Reaching high and stable wheat yields is closely linked to weed control. Herbicide applications to cereal crops in Tunisia have registered a constant increase especially in wet years. In fact, when water conditions are favorable, in order to ensure the best results, the use of herbicides will remove weed competition towards the wheat crop. In addition, foliar herbicides efficiency requires a good humidity of the air (> 60%) and mild temperatures (7-18 ºC).

Based on the national pesticides registration data, this increase of herbicide applications was not accompanied by an increase in the number of the active ingredient registered. While the numbers of active ingredients and chemical families registered in wheat are different (26 and 11 respectively), the modes of action (6) are still very weak. Further complicating the fact that weed control is poor in cereals, the herbicides used by farmers are even more limited. Indeed, our surveys have shown that among the 43 products registered; farmers use only 8. Amilcar and Puma Komplete are the two most widely used herbicides for grass control in wheat.

Worldwide, more weed species are resistant to these types of herbicides than to any other mode of action (Heap 2011). For this reason, it is considered that their use constitutes a high resistance risk. Before that, most farmers relied on ACCase-inhibiting herbicides for the control of grass weeds in cereals. However, rye-grass control failure has been observed and the weed evolved resistance to all ACCase inhibitors that has been tested (Gasquez 2000; Souissi et al., 2004). The introduction of the Amilcar and the Atlantis, a combination of two sulfonylureas, is bound to be a short-term solution. Indeed, problems of effectiveness of these products have already been observed in France and the UK (Resistance to Atlantis can be due to ALS target site resistance or caused by enhanced metabolism; EMR causes broad spectrum of resistance across different modes of action) (Moss 2006; Vacher et al, 2007). Moreover, new herbicides, such as the prosulfocarb and pinoxaden, might not be complete solutions to resistance (Moss 2006). In fact, mechanisms of resistance including target site and non-target site mechanisms that have been selected by ACCase inhibiting herbicides such as fenoxaprop confer cross-resistance to pinoxaden (Yu et al 2007; Petit et al 2010).

Herbicide resistance is a very complex phenomenon that involves several mechanisms of resistance, which cross-resistance patterns could be in some cases very unpredictable and involving a wide range of herbicides and even not yet used. As a result, in the context of lack of innovation, a herbicide becoming ineffective because of resistance is nearly impossible to replace, and each herbicide molecule available is very valuable and should therefore be used as long as possible with a maximum of efficiency. However, in general, an exclusively chemical weeding solution does not seem sustainable in the long term. The implementation of integrated weed management practices will increasingly need to complement chemical means (Chauvel et al 1998).

In Tunisia, a reasonable management of the use of available herbicides by alternating or associating herbicide molecules during crop rotations may be a useful means to reduce the risk of the occurrence of herbicide resistance in weeds, and thus to maintain the effectiveness of these herbicides as long as possible.

Conclusion

Reaching high and stable wheat yields is a national priority in Tunisia. Arable surfaces cannot be increased in a sustainable way. Therefore, wheat crop has become more and more intensive, and chemical weed control is an essential component of productive agriculture.

When considering herbicide use trends in Northern Tunisia, the most important region of wheat production, a turning point is clearly observed around the 1980s. It corresponds to the intensification of the use of selective graminicides. Actually, 85% of sown areas are treated. However, 93% of cereal crops are conducted in rain fed conditions and herbicide applications are closely correlated to rainfall and mainly autumn. Consequently, herbicide-treated areas are subject to year-to-year variation with extremely low values in dry areas. Water, thus, appears to be the first limiting factor of cereal production in Tunisia and hence influencing farmers' decision regardless of herbicide applications.

Graminicides and large spectrum herbicides are the most used products. These herbicides correspond to a very narrow range of modes of action. Their intensive use has led to the development of rye-grass resistant populations. Resistance was confirmed for ACCase and ALS-inhibiting herbicides and mechanisms involved confer cross resistance to a wide range of herbicides even never been used. Chemical weed control remains thus a short-term solution and a rational management of these molecules which is essential to maintain their effectiveness as long as possible. They should be a part of an integrated weed management strategy.

In this study, the usage of herbicides in Northern Tunisia and its relation with the development of resistance is estimated through herbicide- treated areas and the amount of herbicides imported. A large scale survey could be conducted in this region to investigate the relationship between the cultural practices and molecules used with the level of resistance in each farm.

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