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Plant pathology

Effects of adjuvants on herbicidal action. I. Effects of a mixture of adjuvants on diclofop-methyl retention and penetration in wheat and ryegrass

C Gauvrit, JL Dufour

INRA, laboratoire de malherbologie, BV 1540, 21034 Dijon Cedex, France

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Summary — A mixture of adjuvants composed of a liquid nitrogenous fertilizer, oil, solvent and surfactant increases diclofop-methyl efficacy against ryegrass (*Lolium multiflorum* Lam) at application volumes varying from 75 to 500 l•ha^{-1}. The mixture only slightly affects spray retention on ryegrass (whatever the applied volume) and on wheat at 75 l•ha^{-1}, but more than doubles it on wheat at 150, 300 and 500 l•ha^{-1}. Diclofop-methyl penetration is low in wheat (less than 10% in 3 days) as well as through the wettable abaxial surface of ryegrass (less than 5%). By contrast, penetration through the poorly wettable adaxial surface of ryegrass is 29% and 52% at 75 and 500 l•ha^{-1} respectively. The mixture increases herbicide penetration in both species: 2 to 3 times in ryegrass and 4 to 11 times in wheat, according to application volume. Oil plays a major part in this stimulation. Combined effects on retention and penetration lead to an increase in herbicide entry into the plants, which explains the higher efficacy. The increase is 3 to 4 times greater in wheat than in ryegrass, and this tends to diminish selectivity.

diclofop-methyl / adjuvant / retention / penetration / efficacy

Résumé — Effets des adjuvants sur l'action herbicide. I. Effet d'un mélange d'adjuvants sur la rétention et la pénétration du diclofop-méthyl chez le blé et le ray-grass. *Un mélange d'adjuvants contenant de l'engrais azoté liquide, une huile, un solvant et un mouillant améliore l'efficacité du diclofop-méthyl sur ray-grass (* Lolium multiflorum *Lam) pour des volumes d'application variant de 75 à 500 l•ha^{-1} (fig 1). Le mélange d'adjuvants affecte peu la rétention de la pulvérisation par le ray-grass, quel que soit le volume d'application, et par le blé à 75 l•ha^{-1}; par contre il la double pour le blé à 150, 300 et 500 l•ha^{-1} (fig 2). Le diclofop-méthyl pénètre mal chez le blé (moins de 10% en 3 j, fig 4) ainsi que par la face abaxiale (mouillable) du ray-grass (moins de 5%, fig 3A). Par contre, la pénétration est 29% (à 75 l•ha^{-1}) et 52% (à 500 l•ha^{-1}) par la face adaxiale (peu mouillable) (fig 3B). Le mélange d'adjuvants stimule fortement la pénétration de l'herbicide dans les 2 plantes, d'un facteur 2 à 3 chez le ray-grass et 4 à 11 chez le blé, selon le volume appliqué (figs 3 et 4). L'huile joue le rôle essentiel dans cette augmentation (fig 5). Il résulte de ces effets une augmentation de la quantité d'herbicide entrant dans les plantes, ce qui explique l'amélioration d'efficacité. Cet accroissement est 3–4 fois plus important chez le blé que chez le ray-grass, ce qui tend à diminuer la sélectivité de l'herbicide.*

diclofop-méthyl / adjuvant / rétention / pénétration / efficacité

INTRODUCTION

During the last few years the use of adjuvants in herbicide applications has gained considerable popularity among French farmers. It is often associated with a lower treatment volume and herbicide dosage, as well as an earlier application time. The most commonly used adjuvant is a complex mixture (see *Materials and Methods*) comprising mineral oil, surfactant, solvent and liquid nitrogenous fertilizer. The benefits of this technique are currently under debate. Few question the increase in herbicide efficacy against weeds; however doubts have been raised about its innocuity on crops. For example, in field experiments, Orlando and Jouy (1990) found that the mixture of adjuvants brought about phytotoxicity symptoms in wheat (*Triticum sativum* L) treated with diclofop-methyl. In contrast, Bouchet and Beaufreton (1988), while observing increased efficacy of diclofop-methyl and isoproturon against ryegrass (*Lolium multiflorum* Lam), wild-oat (*Avena fatua* L) and blackgrass (*Alopecurus myosuroides* Huds) found no injury to wheat. A reason for this discrepancy could be that environmental conditions play a great part in the appearance of injury in crops. Field trials are underway to elucidate this point.

To our knowledge, no report on the action of such complex mixtures has yet been published but the action of individual components has been well documented. Oils increase droplet spreading on plants (Mc Whorter and Barrentine, 1988; Wanamarta *et al,* 1989; Schott *et al,* 1990) and herbicide penetration (Gillepsie *et al,* 1988; Grafstrom and Nalewaja, 1988; Mc Call, 1988; Wanamarta *et al,* 1989; Gauvrit and Dufour, 1990; Schott *et al,* 1990). Wetting agents increase spray retention (Harper and Appleby, 1984; O'Donovan *et al,* 1985; Anderson *et al,* 1987; De Ruiter and Uffing, 1988) and in some instances penetration as well (Sharma *et al,* 1978; Harper and Appleby, 1984; O'Donovan *et al,* 1985; De Ruiter and Uffing, 1988). Solvents help to impede active ingredient precipitation when it is close to its solubility limit (Galoux *et al,* 1986), thus maintaining it in the physical state most suitable for passage through the cuticle. Liquid nitrogenous fertilizers are hygroscopic (Norden, 1988) and prevent the deposit from drying completely, which is thought to promote penetration (Stevens *et al,* 1988).

We have sought to establish what effect the mixture of adjuvants has on two important efficacy parameters, namely retention and penetration, when diclofop-methyl is applied on ryegrass and wheat, a weed control situation commonly encountered in France.

MATERIALS AND METHODS

Plant material

Wheat (*Triticum sativum* L, cv Pernel) and ryegrass (*Lolium multiflorum* L, cv Adret) seeds were germinated at 25°C and sown in a clay loam soil: sand mixture (1:1). Ryegrass plants were then placed in a growth cabinet at 14/9°C (day/night), 14 h photoperiod and 65/95% relative humidity. Wheat plants were placed in the same conditions except for day temperature: 17°C. All experiments were performed when the third leaf was 1 to 3 cm long.

Diclofop-methyl efficacy on ryegrass

Diclofop-methyl was applied as an Illoxan CE emulsion. Illoxan CE is an emulsifiable solution which contains 36% diclofop-methyl, 49% aromatic solvents and 15% anionic emulsifiers. The emulsion was applied by means of an indoor sprayer consisting of a movable boom with two "Albuz" 110° nozzles positioned 50 cm apart: grey nozzles operated at 4.0 bars for 300 and 500 $l{\bullet}ha^{-1}$, blue nozzles operated at 3.0 bars for 150 $l{\bullet}ha^{-1}$, red nozzles operated at 2.0 bars for 75 $l{\bullet}ha^{-1}$. Plants were placed 48 cm under the nozzles and were sprayed with 150 $g{\bullet}ha^{-1}$ diclofop-methyl. From the mixture compositions commonly used by farmers, we chose the following one. Solvent: 0.5 $l{\bullet}ha^{-1}$ isophorone (Prolabo); surfactant (non-ionic): 0.1 $l{\bullet}ha^{-1}$ Citowett (100% alkyl-aryl phenol polyglycol ether (5 ethylene oxide), BASF Co); oil: 0.5 $l{\bullet}ha^{-1}$ Végélux (840 $g{\bullet}l^{-1}$ emulsifiable mineral oil, CCL Co); 5 $l{\bullet}ha^{-1}$ liquid nitrogenous fertilizer (39% ammonium nitrate, 39% urea). The oil emulsifier was occasionally tested at a 0.12 $l{\bullet}ha^{-1}$ dose.

At the stage defined above, plants were sprayed at the chosen volume, with or without the mixture of adjuvants. Nine replications with 8 plants each were carried out for each treatment and after 14 d under the growth conditions the shoots were cut off at ground level and placed at 80 °C for 24 h for dry weight determination.

Retention measurements

Plants were sprayed under the same conditions as for the efficacy experiments except that diclofop-methyl dosage was 1080 $g{\bullet}ha^{-1}$. The sprays contained 0.01% fluorescein as in Richardson's experiments (1984). After the spray had dried on the foliage, the plants were cut off at ground level and shaken for 30 s in 50 ml 5 mM NaOH. Readings were made in a Jobin and Yvon 3-D spectrofluorimeter at 490/510 nm. Plants were then placed at 80 °C for 24 h and the dry matter weighed. For ryegrass, experiments comprised 6 repetitions with 25 plants each and for wheat, 3 repetitions with 10 plants each.

Diclofop-methyl penetration

Ring ^{14}C labelled diclofop-methyl (257 $MBq{\bullet}mmol^{-1}$, 98.5% radiochemical purity) was dissolved in ethanol. An aliquot containing the desired radioactivity was deposited at the bottom of a conical tube and the ethanol evaporated to dryness. An aqueous emulsion of Illoxan CE was then added at a concentration corresponding to a 1080 $g{\bullet}ha^{-1}$ diclofop-methyl, at either a 75 or 500 $l{\bullet}ha^{-1}$ treatment. Gentle shaking for 2 h interspersed with two 10 s sonication spells redissolved radiolabelled diclofop-methyl. Radioactivity of the preparation was 16.7 $Bq{\bullet}\mu l^{-1}$ and cold herbicide was 99.0% (500 $l{\bullet}ha^{-1}$) or 99.8% (75 $l{\bullet}ha^{-1}$) of total herbicide. Adjuvant concentrations in the applied emulsion corresponded to that defined above and were calculated on a ha basis. It follows that adjuvant concentrations were 6.7 times higher in conditions corresponding to 75 $l{\bullet}ha^{-1}$ treatments than in 500 $l{\bullet}ha^{-1}$ treatments.

Four 0.5-µl droplets of the above emulsion were deposited on the upper third of the 2nd leaf and the plants placed in a growth cabinet at 17/9°C (day/night), 14 h photoperiod and 65/95% relative humidity. Penetration was studied on the adaxial surface of wheat and on both abaxial and adaxial surfaces of ryegrass. Absorption was determined 1 and 3 d after treatment. Each measurement was made on 5 plants from the same pot and the experiment was conducted with 3 replicates. Absorption was evaluated by washing the treated area of each leaf with 1 ml acetone and then 1 ml of chloroform. Washes were combined and evaporated to dryness. Ethanol was added to dissolve diclofop-methyl which was counted in Dynagel (JT Baker Chemicals, The Netherlands) by scintillation counting. Leaves were dried (24 h, 80°C) and combusted in an oxidizer for radioactivity assessment. Recovery figures varied from 81 to 104%.

Statistics

The experiments were full factorial (efficacy and retention studies: 2 factors; penetration studies: 3 factors). Data were submitted to analysis of variance and means were compared using the Newman–Keuls test at the 5% level (Cochran and Cox, 1968). Data marked with the same letters in the figures and tables do not differ significantly.

RESULTS

Diclofop-methyl efficacy on ryegrass

Efficacy was higher at 150 to 500 l•ha^{-1} than at 75 l•ha^{-1} (fig 1). The mixture of adjuvants improved diclofop-methyl efficacy by 11 to 21%. There was no volume x adjuvant interaction.

Retention

The retention of Illoxan CE sprays was higher on ryegrass than on wheat: 2.2 times at 75 l•ha^{-1} and 4.9 times at 500 l•ha^{-1} (fig 2). Retention was roughly proportional to the applied volume on ryegrass but such a relationship was not observed on wheat. Another difference between the two plants was that on ryegrass, retention was not affected by the mixture of adjuvants whereas in wheat it was increased at 150, 300 and 500 l•ha^{-1}. Statistical analysis revealed a volume x adjuvant interaction, the adjuvant effect being all the more important the higher the volume.

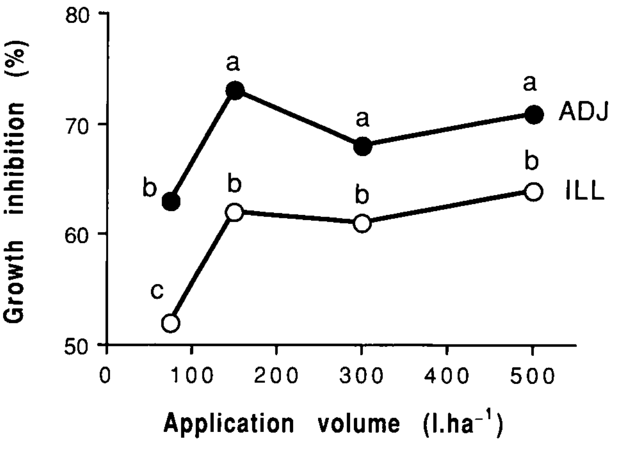

Fig 1. Effect of diclofop-methyl on ryegrass growth as influenced by the mixture of adjuvants and the application volume. Growth was defined as the dry matter increase between treatment and harvest. Growth inhibition is expressed as % control growth (1.563 ± 0.141 g dry matter). ILL: treatment with an Illoxan CE emulsion; ADJ: treatment with an Illoxan CE emulsion combined with the mixture of adjuvants.

The increase in retention on wheat could not be attributed to a single adjuvant (table I).

Penetration

Diclofop-methyl penetration through the abaxial surface of ryegrass was low: after 3 days it amounted to less than 3% of the applied radiolabelled diclofop-methyl at 75 l•ha^{-1} and 5.3% at 500 l•ha^{-1} (fig 3A). The mixture of adjuvants increased penetration to 11.7% and 8.5% at 75 and 500 l•ha^{-1} respectively. There was no volume effect but a volume x adjuvant, interaction; adjuvant effect being lower at high volume.

Table I. Retention of a spray of Illoxan CE emulsion on wheat as influenced by adjuvants.

Adjuvant	*Retention** *(µl•g^{-1} dry matter)*
None	59.6^{a}
Liquid nitrogenous fertilizer	68.9^{a}
Emulsifer	42.8^{a}
Oil	49.5^{a}
Solvent	60.8^{a}
Wetting agent	47.8^{a}
Mixture of adjuvants	90.0^{b}

* Values followed by the same letter do not differ significantly.

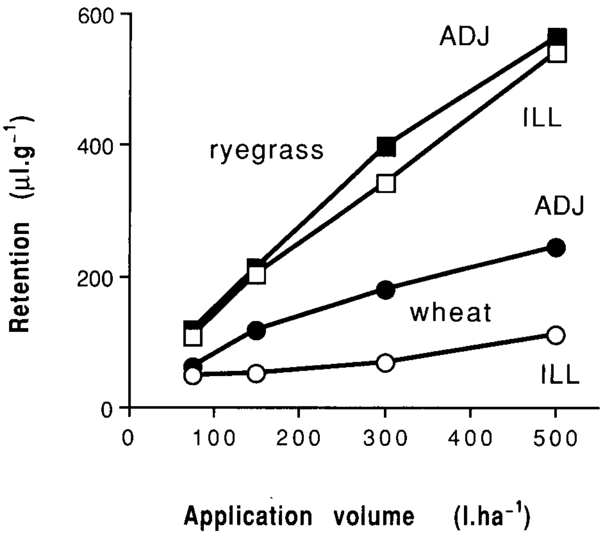

Fig 2. Effect of the mixture of adjuvants on spray retention by ryegrass and wheat. ILL, ADJ: (see fig 1).

After 3 days, diclofop-methyl penetration through the adaxial surface of ryegrass amounted to 28.7% and 51.8% at 75 and 500 l•ha^{-1} respectively (fig 3B). The mixture of adjuvants increased it to 80.0% (75 l•ha^{-1}) and 92.2% (500 l•ha^{-1}). Penetration was significantly higher at 500 l•ha^{-1} but there was no volume x adjuvant interaction.

When diclofop-methyl was administered in the commercial preparation, its penetration into wheat was low: 5.7% at 75 l•ha^{-1} and 10.5% at 500 l•ha^{-1} after 3 days (fig 4). The mixture of adjuvants dramatically increased penetration to 65.0% and 41.8% respectively. The effect was stronger at 75 l•ha^{-1}.

When added individually to Illoxan CE emulsion, adjuvants exhibited different effects on diclofop-methyl penetration (fig 5). The emulsifier, Citowett and isophoron had no significant effect. Liquid nitrogenous fertilizer and Végélux oil improved penetration, the oil effect not being significantly different from that of the complete mixture.

DISCUSSION

Diclofop-methyl efficacy on ryegrass

Diclofop-methyl efficacy is lower at 75 l•ha^{-1} than at 150 to 500 l•ha^{-1} (fig 1). At high volumes, concentration of the active ingredient is low but foliage coverage is good; however, at lower volumes the herbicide concentration is high, at the

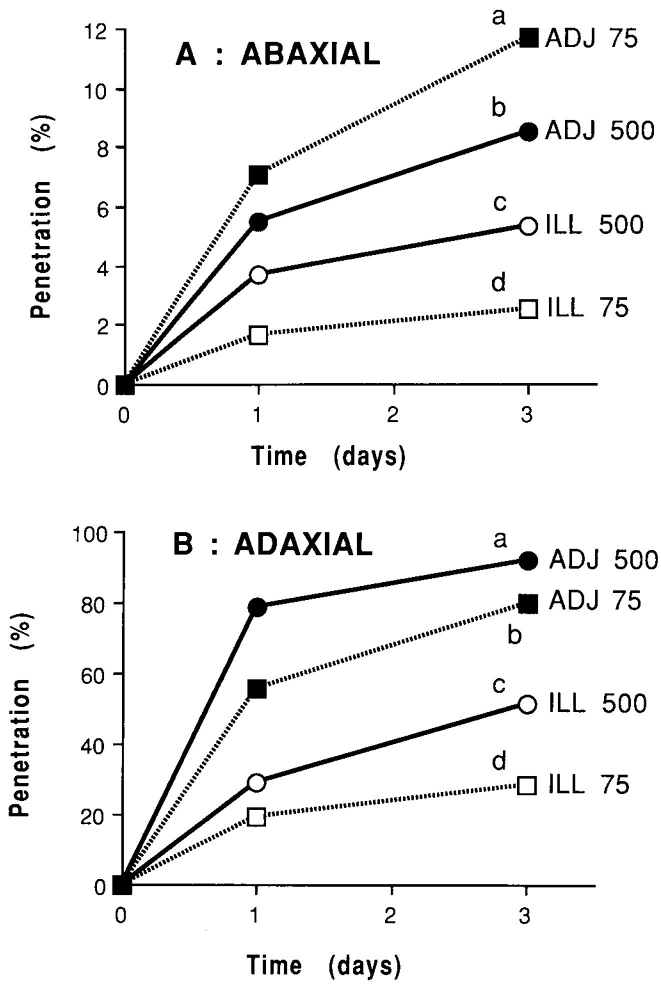

Fig 3. Diclofop-methyl penetration through the abaxial (A) and adaxial (B) surfaces of ryegrass as influenced by the mixture of adjuvants and the application volume. ILL, ADJ: (see fig 1), 75, 500 : l.ha–1. Note scale difference on the penetration axis.

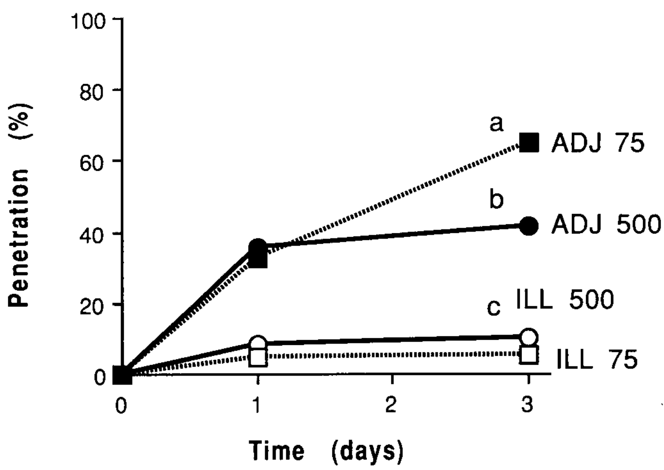

Fig 4. Diclofop-methyl penetration through the adaxial surface of wheat. ILL, ADJ, 75, 500: (see fig 3).

expense of foliage coverage. A few herbicides are more active at high concentrations, for instance glyphosate (Ambach and Ashford, 1982; Merritt, 1982; Kudsk, 1988) and phenmedipham

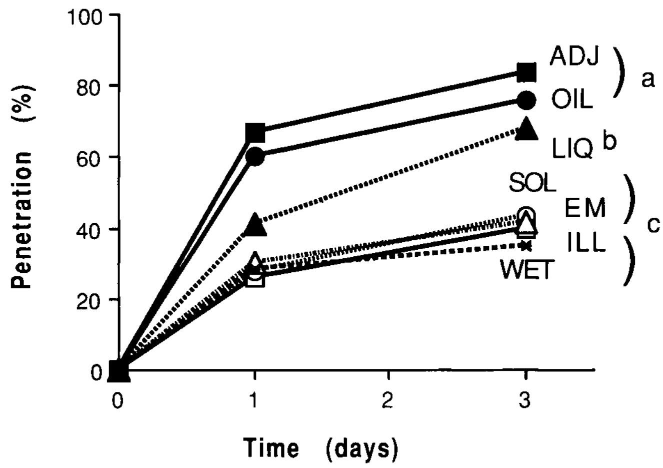

Fig 5. Diclofop-methyl penetration through the adaxial surface of ryegrass as influenced by adjuvants. ADJ, ■ : mixture of adjuvants; EM, Δ: emulsifer; ILL,□: Illoxan; LIQ, ▲ : liquid nitrogenous fertilizer; OIL, ●: oil; SOL, O: solvent; WET, X: wetting agent

(Bishof, 1988). In contrast, contact herbicides are generally thought to require high application volumes to exert maximal action (Taylor, 1981), but this opinion is not shared by Skuterud *et al* (1988). Our results show that diclofop-methyl falls into the second category, although its action cannot be qualified as contact. It must be stressed that since we used different nozzles to deliver the various application volumes, differences in droplet spectra might have played a part in the observed differences in efficacy.

The mixture of adjuvants improved diclofop-methyl efficacy by 11 to 21%. This increase was somewhat less extensive than that observed by Bouchet and Beaufreton (1988). However, these authors had measured diclofop-methyl efficacy 30 and 60 days after treatment, whereas we made our observations 14 days after treatment. As diclofop-methyl is a slow acting herbicide, this could explain why our results are qualitatively but not quantitatively in agreement.

Retention

As shown by Schott *et al* (1990) the high wettability of *Lolium multiflorum* is due to the exposure of the abaxial surfaces towards the spray at the 3-leaf stage. Diclofop-methyl spray retention on wheat is noticeably lower, which is in agreement with the selectivity of this herbicide.

It is also worth noting that in ryegrass, retention is roughly proportional to the applied volume whereas such a relationship is not found in wheat. From the shape of the two curves (fig 2) it follows that lowering the applied volume at constant dosage per ha does not alter the amount of active ingredient collected by ryegrass but increases it in wheat. For example, whilst retention by wheat is divided by 2.2 when the volume is lowered from 500 to 75 l•ha^{-1}, collection of the active ingredient is increased by 3.0.

The two preceding remarks show that when diclofop-methyl is applied to wheat to control ryegrass, volume reduction tends to lower selectivity. However, the predominant selectivity factor in this case is diclofop-methyl degradation by wheat (Shimabukuro *et al*, 1979; Boldt and Putnam, 1981).

In ryegrass we observed no increase in retention upon addition of the mixture of adjuvants to the Illoxan CE emulsion. This is in line with observations by Blackman *et al* (1958) and De Ruiter and Uffing (1988) who found that on wettable plants the increase in retention by surfactants was either nil or limited. However, in wheat the mixture of adjuvants increased retention (fig 2). The effect was not noticeable at 75 l•ha^{-1}, possibly because at such a volume the concentration of the adjuvants already present in the commercial preparation is high. Surface tension of Illoxan CE emulsions is low: 30 to 31 mN•m^{-1} (Schott *et al*, 1990), indicating the presence of tensio-active substances. Their concentration may be so high at 75 l•ha^{-1} that further addition of surfactants through the mixture of adjuvants does not improve their action on retention. This explanation is in line with the shape of the retention curve for wheat, which levels off as the applied volume decreases.

Since the increase in retention cannot be attributed to a single adjuvant (table I), we can hypothesize that it is the result of a synergy between two or more components of the mixture, but so far we have not carried out any experiments to verify this idea.

Penetration

The two ryegrass (*Lolium perenne* L) leaf surfaces have different properties with regard to wettability (Field and Bishop, 1988). Contact angles of formulated glyphosate droplets are 36° and 118° for the abaxial and the adaxial surface respectively. The latter is covered with dense deposits of crystalline wax whereas the abaxial surface presents amorphous epicuticular waxes. We found the same wettability difference between the two leaf surfaces of *Lolium multiflorum* (Schott *et al*, 1990). In addition, we have shown that diclofop-methyl penetration is one order of magnitude higher on the adaxial surface. This is reminiscent of Holloway and Silcox's observations (1985) that non-ionic surfactant penetration is faster through waxy leaves such as rape *(Brassica napus* L) and pea *(Pisum sativum* L) which possess microcrystalline wax deposits on the surface.

In the absence of adjuvants, diclofop-methyl penetration was always higher at 500 $l \cdot ha^{-1}$ than at 75 $l \cdot ha^{-1}$, which might explain why efficacy on ryegrass was lower at 75 $l \cdot ha^{-1}$ than at 500 $l \cdot ha^{-1}$ (fig 1). It also shows that, under our conditions, increasing the concentrations of the active ingredient and adjuvants does not promote penetration.

The mixture of adjuvants drastically increases diclofop-methyl penetration in ryegrass (fig 3) which very probably explains the improvement in herbicidal efficiency upon their addition. However, the beneficial effect of high volumes which was observed in biological tests (fig 1) was found only on the adaxial surface with regard to penetration.

All adjuvants present in the mixture do not participate equally in the increase in penetration. The solvent (isophoron), the emulsifier and the surfactant (Citowett) have no detectable effect.

Isophorone's lack of action is understandable since diclofop-methyl is in a dissolved state in the emulsion. Both the emulsifier and Citowett could be expected to increase diclofop-methyl penetration since surfactants can promote herbicide entry into plants (Sharma *et al*, 1978; Harper and Appleby, 1984; O'Donovan *et al*, 1985). However, since Illoxan CE is an emulsion, there is an emulsifier present in the commercial preparation. This might be the reason why further addition of this type of compound does not affect diclofop-methyl penetration.

Liquid nitrogenous fertilizer increases diclofop-methyl penetration, as it increases the efficacy of some herbicides (Horn *et al*, 1986; Chow, 1988), but not all (Sander *et al*, 1987). Liquid nitrogenous fertilizer is hygroscopic (Norden, 1988) and maintains an aqueous environment inside the deposit. It favours penetration (Stevens *et al*, 1988) since the activity and the diffusion of the active ingredient are maintained (Price, 1982). We observed that in the presence of the mixture of adjuvants the deposits never took on the appearance of a white precipitate as was the case with Illoxan CE emulsion alone. However, Babiker and Duncan (1975) have shown that humectants such as glycerol and sorbitol reduce aminotriazole penetration into bracken *(Pteridium aquilinum* L).

The action of oils is poorly understood, even though numerous reports show that they promote herbicide entry into plants (Gillepsie *et al*, 1988; Grafstrom and Nalewaja, 1988; McCall, 1988; Wanamarta *et al*, 1989; Gauvrit and Dufour, 1990; Schott *et al*, 1990). The fact that the addition of oil brings about the same stimulation of penetration as the complete mixture of adjuvants, together with the lack of effect of the mixture on retention by ryegrass, confirms outdoor results that oil also plays the predominant role in the increase in efficacy (Bouchet and Beaufreton, 1988). It also indicates that the mixture might be too complicated for its purpose.

Combining the effects on retention and penetration it follows that at 75 $l \cdot ha^{-1}$ the mixture of adjuvants increases 3 to 4 times the amount of diclofop-methyl that enters ryegrass, whereas the corresponding figure for wheat is 11.5. At 500 $l \cdot ha^{-1}$ these figures are 2 and 8 respectively. Hence, the mixture of adjuvants increases herbicide entry into plants more in wheat than in ryegrass, which is adverse to selectivity. In this respect, field trials have shown that, even at reduced diclofop-methyl dosage, oil addition can cause injury to wheat (Orlando and Jouy, 1990). However, Bouchet and Beaufreton (1988) in outdoor experiments with the same mixture of adjuvants showed increased activity against ryegrass and a lack of phytotoxicity against wheat. Hence, the increase of herbicide entry in wheat only produces injury under certain conditions which remain to be defined.

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