JOURNAL OF
**AGRICULTURAL AND
FOOD CHEMISTRY**
A R T I C L E

Effect of a Spreading Adjuvant on Mesotrione Photolysis on Wax Films

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There is little information about pesticide photostability on plants, especially when considering the effects of the formulation. We evaluated the photostability of a herbicide, mesotrione, on wax films. These surfaces are good systems to mimic the outer layer of the leaf. Within the range of recommended agricultural rates, pure mesotrione half-life on cuticular wax films was between 100 and 160 min. Formulated, the phototransformation rate was multiplied by a factor of 4.8. We assume that the acceleration is mainly due to the surfactants, agents that allow a better spreading of the active ingredient at the leaf surface. Since mesotrione photolysis is a fast process on wax films, we can assume that this process would be significant in the field after treatment.

KEYWORDS: Phototransformation; cuticular wax; leaves

INTRODUCTION

The objective of this research was to determine to what extent adjuvants for agrochemicals interact with the active ingredient of photochemistry at the leaf surface. Pesticide reactivity with respect to sunlight after crop treatment is seldom evaluated on plant leaves (*1*). Moreover, the effect of adjuvants is rarely considered (*2*). This study aims to point out the possible importance of photochemical reactions on crops after pesticide application and the need to adjust the laboratory experimental setup to field conditions (applied dose, use of adjuvants, etc.).

Foliar application of pesticides is a critical procedure in modern agriculture (*3*). Indeed, the success of a given phytosanitary treatment depends on whether the active ingredient reaches its biological target. At this stage, adjuvants are essential (*4*). They allow a better retention of the spray on the plant and, once there, better resistance to environmental factors (wind, rain, dew formation, and exposure to UV radiation). Numerous studies have described the effect of surfactants on active ingredient diffusion through plant cuticles. Forster et al. showed a correlation between surfactant concentration per unit area and uptake of the active ingredient (*5*). Research studies on adjuvants designed to control volatilization (*6*) or rainfastness (*7*) are under way. However, there is little information about the effect of adjuvants on pesticide photostability on plants.

The function and physical and chemical properties of an active ingredient largely determine the type of adjuvant system (*8*). We have focused the discussion here on herbicides as they dominate the adjuvant market. Surfactants are the most important class of adjuvants for herbicides; they are also called surface active agents. They reduce the surface tension of the spray solution to allow a

more intimate contact between the droplet and the leaf surface (*9*). This results in a better absorption of the herbicide. In addition, surfactants may also promote herbicide diffusion through the cuticle. Several studies showed an effect of surfactants on active ingredient photolysis in water (*10*, *11*) or acetonitrile (*12*). On wax films, which are surfaces mimicking the outer layer of the leaf, formulated sulcotrione photodegrades faster than the pure active ingredient (*13*). However, to our knowledge no studies have ever tried to understand the effects of surfactants on pesticide photolysis on plant leaves.

Two main kinds of surfactant are usually described: aryl and alkyl ethoxylated surfactants. There is a trend to abandon aryl surfactants due to their endocrine disrupting effects. Among the alkyl ethoxylated surfactants, a wide range of chemical structures have been developed (*14*). For this study we selected a combination of herbicide and alkyl ethoxylated surfactant that is encountered among commercial formulations. We chose mesotrione as the herbicide, which is a relatively new active ingredient used for maize treatment (**Figure 1**). Callisto is one of its widely used commercial formulations. As commercial formulation recipes are usually secret, we only found some information about it in the security data sheet, which states that it contains an alkyl ethoxylated surfactant (ethoxylated isodecyl ether). The ratio between the amount of surfactant and the amount of active ingredient is around 5.

We have seen elsewhere that the commercial formulation of mesotrione promotes mesotrione photolysis on wax films (*15*). Here, we discus the possible means of adjuvant interaction with mesotrione photolysis. We also consider the effect of the commercial formulation or the alkyl ethoxylated surfactant alone.

EXPERIMENTAL SECTION

Chemicals. All solvents and chemicals were used as received. Mesotrione (99.9%) was purchased from Riedel de Haën (Saint-Quentin

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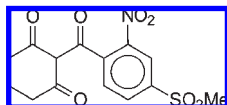
Figure 1. Mesotrione structure.

Fallavier, France). Mesotrione commercial formulation, Callisto (100 g/L of mesotrione) was obtained from a regular agricultural shop. The alcohol ethoxylate used was isodecyl alcohol polyethylene glycol (CAS No. 61827-42-7); the acronym used is PEO-10-6 where 10 refers to the length of the fatty alcohol and 6 to the average number of ethylene oxide units. Methylene chloride (gradient grade) and acetonitrile (HPLC grade) were obtained from Riedel de Haën (Saint-Quentin Fallavier, France). Formic acid (99−100%), potassium dihydrogen-ophosphate (99.5%), and disodium hydrogenophosphate (99%) were obtained from Prolabo (VWR, Fontenay sous Bois, France). Water was purified using a Millipore milli-Q system (Millipore Q, resistivity 18 MΩcm, DOC < 0.1 mg/L). For sunlight actinometry 4-nitroanisole (97%) was supplied by Aldrich (Saint-Quentin Fallavier, France) and pyridine (99%) by Lancaster (Alfa aesar, Schiltingheim, France).

Analytical Methods. The HPLC-UV analyses were performed at room temperature, using a Waters 2487 dual wavelength detector. The detection wavelength was set to 270 nm. The reverse phase column had the following characteristics: RP18 5 μm, 250 mm × 4.6 mm X-terra (Waters). A 25 μL injection of each sample was made in duplicate using an autosampler. The mobile phase consisted of 35% MeOH and 65% water acidified at pH 2.5 with 3 ‰ of formic acid. A flow rate of 1 mL/min was used for all analyses. The temperature was set to 25 °C for the column and the autosampler compartments.

Photoreaction Setup. Photochemical experiments were conducted in a Suntest CPS photosimulator (Atlas) equipped with a Xe lamp and a special glass filter restricting the transmission of wavelengths below 290 nm. The lamp was set to maximum intensity (765 W/m^2) corresponding to midday sunlight intensity in summer time at a latitude of around 25° N. Cold water flowed through the bottom of the photosimulator to maintain the internal temperature at approximately 27 °C. Glass dishes coated with maize wax films and treated with the herbicide were randomly arranged in the Suntest reactor. Samples were covered by a quartz plate to protect them from the air cooling system. Nitroanisol (PNA) and pyridine were the chemical actinometers used in this study to monitor any variation in light intensity (*16*). The intensity of the light emitted from the Suntest reactor was homogeneous over the 20 positions in the system and constant over the duration of the study.

Film Preparation. The procedure for maize wax extraction and film preparation has been described elsewhere (*13*).

Irradiation on Wax Films. After film preparation, 3 mL of aqueous mesotrione was added on the films. The solutions contained either pure mesotrione, formulated mesotrione, or mesotrione mixed with the surfactant PEO-10-6. Water was evaporated under a fan overnight. The resulting mesotrione concentration per unit area varied from 110 up to 1700 g/ha. Values are specified in the Results section. After irradiation, films were rinsed with phosphate-buffered solution, and samples were analyzed by HPLC. For each irradiation time, two samples were prepared. HPLC analyses were made in duplicate. Control samples in the dark were run concomitantly.

Data Processing. Mesotrione concentration was calculated from the HPLC results and plotted as ln(C/C_0) as a function of irradiation time. Data points were adjusted and pseudo-first-order rates were calculated. Relative error on rate was determined with Student's test.

Comparison between two rates was based on a statistical hypothesis test with a critical region of 5%: if the rates were significantly different, we reported the *p*-value.

IRTF Microscopy. Mesotrione deposits on films were monitored using infrared spectrometry. IR spectra were recorded using a Thermo Scientific Nicolet 6700 spectrophotometer (nominal resolution of 4 cm^{-1}; 64 scan summation). This apparatus was equipped with a Nicolet continuum XL microscope allowing video image capturing and micro-FTIR measurements by reflection (displacement: 5 or 10 μm, window width: 10 to 15 μm).
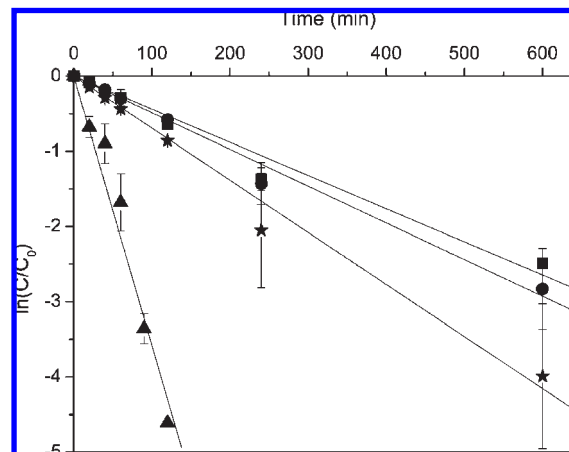

Figure 2. Photolysis of formulated mesotrione (Callisto) on maize wax films at different concentrations: ■, 75 g/ha; ●, 100 g/ha; ★, 150 g/ha; and ▲, 1700 g/ha.

Table 1. Mesotrione Pseudo-First-Order Photolysis Rate Constants

compound or mixture applied	mesotrione concentration	k (s^{-1})	error (%)
pure mesotrione	110 g/ha	2.5×10^{-5}	25%
pure mesotrione	1700 g/ha	3.1×10^{-5}	20%
formulated mesotrione	75 g/ha	7.3×10^{-5}	15%
formulated mesotrione	110 g/ha	8.1×10^{-5}	15%
formulated mesotrione	150 g/ha	1.2×10^{-4}	20%
formulated mesotrione	1700 g/ha	4.6×10^{-4}	25%
mesotrione + PEO-10-6 (R = 0.5)	110 g/ha	3.9×10^{-5}	10%
mesotrione + PEO-10-6 (R = 5)	110 g/ha	1.0×10^{-4}	20%
mesotrione + PEO-10-6 (R = 50)	110 g/ha	1.2×10^{-4}	10%

RESULTS AND DISCUSSION

Recommended agricultural doses for Callisto application vary from 75 g/ha up to 150 g/ha. We first designed an experiment to observe the effect of the applied dose on the rate of mesotrione photolysis on maize wax films. Several amounts of formulated mesotrione corresponding to 75, 110, 150, and 1700 g/ha were tested. The dose of 1700 g/ha is well above the recommended agricultural dose; it was considered in order to open the discussion about the laboratory experimental set up. We did not record any loss of mesotrione in the darkness. We can conclude that formulated mesotrione did not diffuse into the wax films during the time course of the experiment and that there were no volatilization, no thermal degradation, and no biodegradation. When exposed to light, mesotrione decay takes place through photolysis only (see **Figure 2**). The pseudo-first-order constants and corresponding errors (95% confidant bands) are listed in **Table 1**.

The photolysis rates of formulated mesotrione at 75 and 110 g/ha were not significantly different while that at 150 g/ha was significantly different and higher (*p*-value < 1.2×10^{-3}). Within the range of recommended agricultural rates, formulated mesotrione half-life on cuticular wax films was between 100 and 160 min. With those low values in mind, we can assume that photolysis would be a significant dissipation path for mesotrione on crops after treatment. At the dose of 1700 g/ha, formulated mesotrione photolysis is even faster. The photolysis rate is multiplied by a factor of 4 compared to the dose of 150 g/ha. It is, thus, crucial to work within the range of recommended agricultural doses when discussing the possible implication of the results in the field.

For pure mesotrione, the effect of concentration on the rate of photolysis was tested with two concentrations: 110 and 1700 g/ha
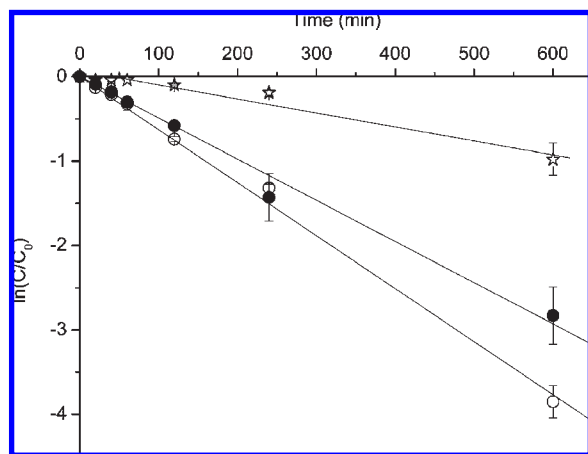
Figure 3. Mesotrione decay under irradiation on wax films (110 g/ha): ☆, pure; ○, mixture with PEO-10-6; and ●, the commercial formulation.

(**Table 1**, lines 1 and 2). The rate constants are not significantly different. Therefore, the acceleration observed with formulated mesotrione is due to the effect of additives contained in the commercial product.

Formulated mesotrione is a complex mixture, whose composition is not accessible. To simplify the system, we considered the simple combination mesotrione plus ethoxylated isodecyl ether (PEO-10-6), which is a main constituent of the formulation. We compared the photolysis rates on maize wax films for pure mesotrione, formulated mesotrione, and mesotrione plus PEO-10-6. Mesotrione concentration was set at 110 g/ha. The amount of PEO-10-6 was set at 550 g/ha, which is five times the amount of mesotrione (the ratio $R = C_{surfactant}/C_{mesotrione}$ is equal to 5). This ratio approaches that of the commercial formulation. Mesotrione phototransformation decay in a mixture with PEO-10-6 is shown on **Figure 3** and the pseudo-first-order rate constants are listed in **Table 1**.

The phototransformation rate of formulated mesotrione was multiplied by a factor of 4.8 compared to that of pure mesotrione. When considering the mesotrione plus PEO-10-6 mixture, the photolysis rate was higher by a factor of 3.9 compared to that of pure mesotrione. If we compare formulated mesotrione and the mesotrione PEO-10-6 mixture, the rates of photolysis are really close. From a photochemical point of view, the commercial formulation is equivalent to the simple mixture with PEO-10-6. We can thus assert that the accelerated phototransformation of mesotrione in Callisto is mainly due to the surfactant. This additive is used to enhance the spreading of the sprayed chemicals on the treated leaves in crops. Here, we showed that this spreading also promotes phototransformation of the active ingredient on wax films. PEO-10-6 did not show any absorption in the actinic portion of solar light. There were also no differences between the absorption spectra of pure mesotrione and the commercial formulation. We thus assume that the accelerating effect of PEO-10-6 or the additives in Callisto (commercial formulation) does not imply any photochemical activity for the additives. The effect is more physical as the additives modify the distribution of mesotrione at the film surfaces and consequently its interaction with light.

Finally, we tried to understand the effect of an increasing amount of PEO-10-6 on mesotrione photolysis. Mesotrione surface concentration was kept constant and equal to 110 g/ha, and the ratio $R = C_{surfactant}/C_{mesotrione}$ was changed. We tested three different ratios: 0.5, 5, and 50. The ratio $R = 5$ corresponds to the ratio in Callisto, which is presented on **Figure 3**. However, to get a complete overview, the curves for the three different ratios were
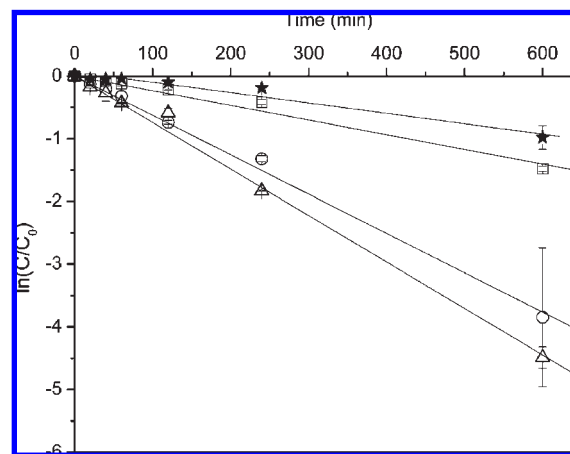

Figure 4. Effect of PEO-10-6 concentration on mesotrione phototransformation (110 g/ha) on maize wax films. ★, pure mesotrione, mixture with PEO-10-6; □, at a ratio $R = 0.5$; ○, $R = 5$; and △, $R = 50$.
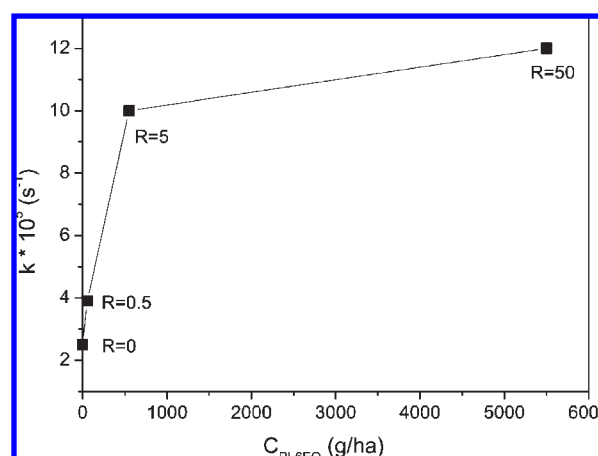

Figure 5. Evolution of the mesotrione photolysis rate as a function of PEO-10-6 concentration per unit area. *R* is the ratio of the amount of PEO-10-6 to the amount of mesotrione.

plotted on **Figure 4** together with the curve obtained with pure mesotrione. By comparing the pseudo-first-order rate listed in **Table 1**, a correlation can be seen between the mesotrione photolysis rate and the amount of surfactant added to the samples. When the amount of surfactant per hectare increases, the mesotrione photolysis rate increases as well. However, the change is not regular.

When plotting the pseudo-first-order constant as a function of the surfactant concentration, two regions appear (**Figure 5**). The photolysis rate increases sharply up to a ratio of 5. Above this value, the increase is much slower. The ratio of 5, which is the ratio reached in the commercial formulation, is the most efficient for mesotrione phototransformation. Above this ratio, a large increase in surfactant concentration leads to a small increase only in the phototransformation rate. Better spreading promoted by a higher amount of surfactants up to the ratio of 5 would promote mesotrione photolysis. Above this value, an additional amount of PEO-10-6 would not promote better spreading, and consequently, only little or no increase in the phototransformation rate can be expected. Similar observations on solution spreading related to surfactant concentration have been made by Pierce et al. (*17*) with a nonionic alkyl polyoxyethylene surfactant. Under certain conditions (high humidity and hydrophilic surface), droplet spreading increases up to a given surfactant amount; above this amount, no further spreading is observed.
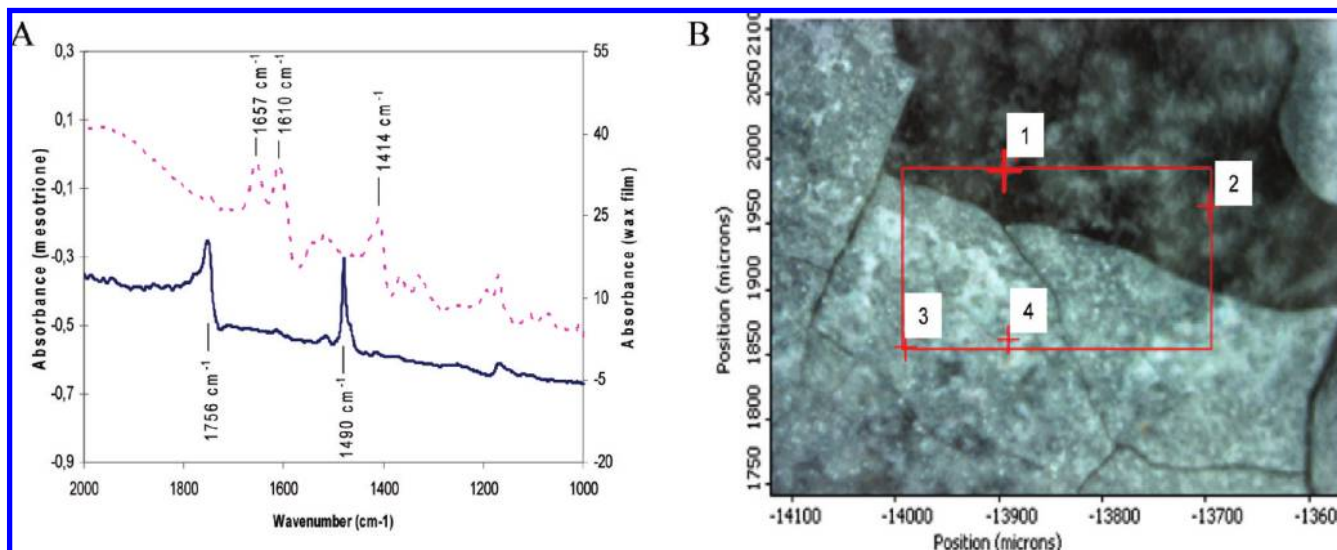
Figure 6. (**A**) IR spectra of (- - -) mesotrione and (—) carnauba gray wax by IR microscopy. (**B**) Microscopic view of a deposit of mesotrione on carnauba wax at the rate of 150 g/ha.
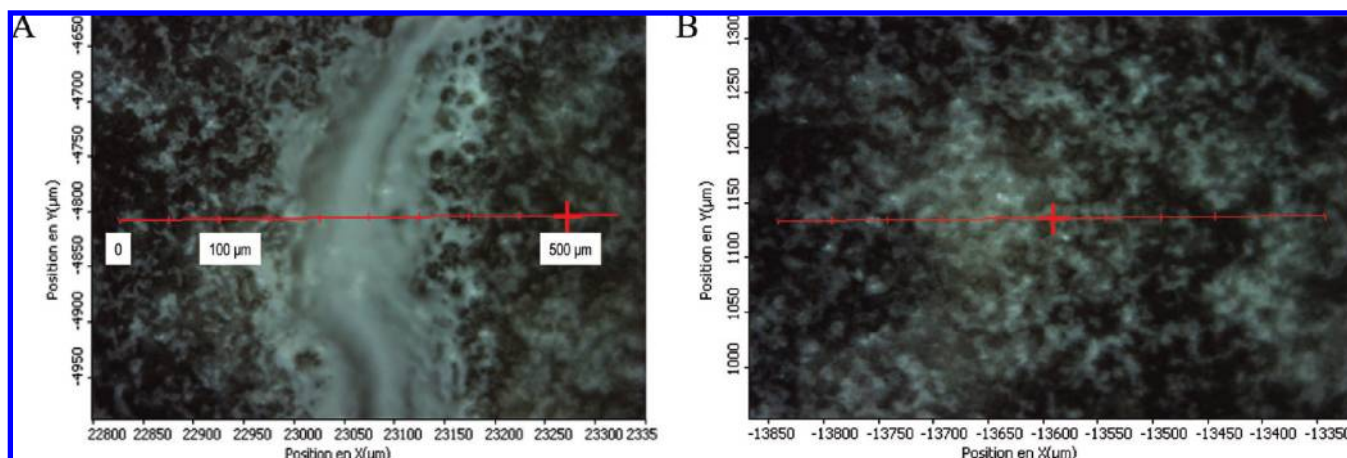

Figure 7. (**A**) Microscopic view of mesotrione deposit on carnauba wax at the rate of 150 g/ha with PEO-10-6 at the ratio $R = C_{surfactant}/C_{mesotrione} = 0.5$. (**B**) Microscopic view of the mesotrione deposit on carnauba wax at the rate of 150 g/ha with PEO-10-6 at the ratio $R = 5$.

To better understand the effect of additives on mesotrione deposits on wax films, we carried out some IR-microscopy analyses. Mesotrione was added on wax films as an aqueous solution. After evaporation of the droplets, a shiny deposit could usually be seen at the surface of the wax films. It contained the active ingredient mixed with additives (*18*) with only a low water content (*5*). Moreover, the distribution of the residues was not homogeneous. Indeed, Pierce et al. recently described the formation of rings and islands of surfactants and active ingredient after droplet evaporation (*10*).

We characterized mesotrione deposits on wax films using IR-microscopy. Pure mesotrione forms small crystals aggregated in well-defined areas on the wax surface. In **Figure 6B**, the crystals were located in the bottom part of the picture, while the wax surface was distinctly in the upper part. By IR, the two areas were well characterized. Positions 1 and 2 on the wax area present the IR spectrum characteristic of the wax (maxima at 1734 and 1472 cm^{-1}) (**Figure 6A**). In the region of 3 and 4, the IR spectra correspond to the pure mesotrione spectrum (**Figure 6A**).

When the surfactant was added, mesotrione deposits changed drastically. At the ratio $R = 0.5$, mesotrione deposits were amorphous. The deposits were still located in a small area in the surface wax; the white deposit in **Figure 7A** is about 150 μm

wide. By IR, we could attribute the white region with no ambiguity to mesotrione and PEO-10-6. For mesotrione, the characteristic bands are at 1660 cm^{-1} and for the alcohol polyethylene glycol at 1490 cm^{-1}.

When the surfactant was added at the ratio $R = 5$, mesotrione deposits were no longer visible with the microscope (**Figure 7B**). The characteristic bands of mesotrione and PEO-10-6 were just discernible in the carnauba wax signal across the surface wax. Mesotrione seemed uniformly distributed at the wax surface.

The drastic change in mesotrione distribution at the wax surface was accompanied by a change in photochemical reactivity. Without or with surfactant mesotrione deposits were crystalline or amorphous, respectively. This first modification should surely result in a modification of light absorption by mesotrione. This could explain the gap between pure and formulated mesotrione reactivity. Second, when the ratio surfactant/additive increased toward a maximum spread of the molecule at the surface, the amount of light absorbed by the mesotrione should also be maximum. This results in maximum photochemical reactivity.

Conclusions. Mesotrione photolysis under simulated solar light on wax film is a fast process. Photodegradation should be a main dissipation factor in the field. Within the recommended

dose range, the photolysis rate of formulated mesotrione is multiplied by a factor of nearly two when considering 75 g/ha and 150 g/ha concentrations. When the concentration of formulated mesotrione is far above this range (1700 g/ha), the photolysis rate is multiplied by a factor of 6. If we want to describe faithfully what happens in the fields, it is very important to remain in the recommended agricultural application dose range.

From this study, we can conclude that the effect of adjuvants on pesticide photolysis on wax film can be very considerable. We have demonstrated here a correlation between mesotrione distribution on a wax film and its photochemical reactivity. Surfactant enhances mesotrione spreading on films, and this is accompanied by faster photolysis. The higher the concentration of surfactant, the faster the photolysis becomes. It would be interesting to perform active ingredient photolysis tests during the elaboration of a formulation in order to optimize the efficacy of the pesticide.

LITERATURE CITED

(1) Pirisi F. M.; Angioni A.; Cabizza M.; Cabras P.; Maccioni E. *J. Agric. Food Chem.* **1998**, *46* (2), 762−765.

(2) Minello, E. V.; Lai, F.; Zonchello, M. T.; Melis, M.; Russo, M.; Cabras, P. *J. Agric. Food Chem.* **2005**, *53* (21), 8302−8305.

(3) Zabkiewicz, J. A. *Weed Res.* **2000**, *40* (1), 139−149.

(4) Green, J. *Pestic. Outlook* **2000**, *11* (5), 196−199.

(5) Forster W. A.; Zabkiewicz J. A.; Riederer M. *Pest Manag. Sci.* **2004**, *60* (11), 1105−1113.

(6) Knowles A. *Pestic. Outlook* **2001**, *12* (5), 182−183.

(7) Spanoghe, P.; Claeys, J.; Pinoy, L.; Steurbaut, W. *Pest Manag. Sci.* **2005**, *61* (8), 793−798.

(8) Ramsey R. J. L.; Stephenson G. R.; Hall J. C. *Pestic. Biochem. Physiol.* **2005**, *82* (2), 162−175.

(9) Sefiane, K. *J. Colloid Interface Sci.* **2004**, *272* (2), 411−419.

(10) Tanaka, F. S.; Wien, R. G.; Mansager, E. R. *J. Agric. Food Chem.* **1979**, *27* (4), 774−779.

(11) Tanaka, F. S.; Wien, R. G.; Mansager, E. R. *J. Agric. Food Chem.* **1981**, *29* (2), 227−230.

(12) Scrano L.; Bufo S. A.; Perucci P.; Meallier P.; Mansour, M. *Pestic. Sci.* **1999**, *55* (9), 955−961.

(13) ter Halle A.; Drncova D.; Richard C. *Environ. Sci. Technol.* **2006**, *40* (9), 2989−2995.

(14) Forster W. A.; Zabkiewicz J. A.; Liu Z. *Pest Manag. Sci.* **2006**, *62* (7), 664−672.

(15) Lavieille, D.; ter Halle, A.; Richard, C. *Environ. Chem.* **2008**, *5* (6), 420−425.

(16) Dulin, D.; Mill, T. *Environ. Sci. Technol.* **1982**, *16* (11), 815−820.

(17) Pierce, S. M.; Chan, K. B.; Zhu, H. *J. Agric. Food Chem.* **2008**, *56* (1), 213−219.

(18) Shi, T.; Schoenherr, J.; Schreiber, L. *J. Agric. Food Chem.* **2005**, *53* (7), 2609−2615.