Studies on Octylphenoxy Surfactants. 7. Effects of Triton X-100 on Sorption of 2-(1-Naphthyl)acetic Acid by Tomato Fruit Cuticles

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The effects of a polyethoxy (EO) derivative of octylphenol (OP) with 9.5 ethyleneoxy groups (OP + 9.5EO) on 2-(1-naphthyl)acetic acid (NAA) sorption by cuticles enzymatically isolated from mature tomato (*Lycopersicon esculentum* Mill. cv. Sprinter) fruit were studied at pH 3.2 and 25 °C. Time to reach NAA sorption equilibrium by cuticular membranes (CM) or dewaxed CM (DCM) was not affected by OP + 9.5EO. At concentrations below the critical micelle concentration (CMC), OP + 9.5EO had little effect on NAA sorption. However, at concentrations above the CMC, NAA sorption was inversely related to OP + 9.5EO concentration. The amounts of NAA sorbed by CM and DCM for the lowest OP + 9.5EO concentration examined (0.001%, w/v) were approximately 50 and 60 μmol·kg^{-1}, respectively. At concentrations above 4%, no detectable amount of NAA was sorbed by CM. Sorption of NAA by CM was inversely related to solution pH (2.2–6.2), and OP + 9.5EO (0.1%) lowered NAA sorption at all pH levels examined. The amount (micromoles per kilogram) of NAA sorbed by CM increased with increasing NAA concentration (micromolar to millimolar). OP + 9.5EO (0.1%) decreased NAA sorption by CM over the entire NAA concentration range examined.

Surfactants, or surface-active agents, are commonly used in agrochemical formulations to improve the performance of an active ingredient (Behrens, 1964; Ford et al., 1965; Foy and Smith, 1969; Parr and Norman, 1965). Possible mechanisms by which this is achieved include stabilization of emulsions and/or suspensions, increased retention of the active ingredient on plant surfaces, and increased wetting of plant surfaces and subsequent penetration of the active ingredient into plant tissues. However, there is evidence that surfactants do not always enhance foliar absorption of an applied compound (Jansen et al., 1961; Temple and Hilton, 1963). This ambiguity has led to several studies examining foliar absorption from specific surfactant/active ingredient combinations (Foy and Smith, 1965; Smith et al., 1966; Stevens and Bukovac, 1987b).

A better understanding of surfactant/active ingredient/plant surface interactions may provide insight into the mechanism(s) of surfactant action. Surfactants are known to alter several physical and chemical properties of the spray solution (Parr and Norman, 1965; Rosen, 1978). It seems clear, however, that these solution properties, as well as surface wetting, are not the sole factors modifying foliar absorption (Foy and Smith, 1965; Hughes and Freed, 1961; Jansen, 1965; Jansen et al., 1961).

The plant cuticle is the initial (Esau, 1977) and primary (Bukovac et al., 1981) barrier to the penetration of foliar-applied compounds. Some attention has been focused previously on the plant cuticle and on interactions between surfactants/active ingredients and this nonliving boundary layer. Smith et al. (1966) suggested that certain polyethoxylated alkylphenol nonionic surfactants orient themselves in cuticular channels or imperfections in a manner that aids subsequent herbicidal penetration. The possibility of surfactant copenetration (Stevens and Bukovac, 1987b) with the active ingredient would indirectly support this view. Numerous reports suggest surfactant solubilization of cuticular lipids as a possible mechanism of surfactant action (Allen, 1979; Foy and Smith, 1969; Furmidge, 1959a,b; Jansen, 1964). However, no conclusive data are available, and there is some evidence suggesting that this is not a universal mechanism of action (Shafer and Bukovac, 1986; Stevens and Bukovac, 1987a).

Use of isolated plant cuticles allows for explicit examination of surfactant/active ingredient/cuticle interactions. Preliminary data suggest this approach is useful (King, 1982; Norris, 1973; Shafer and Bukovac, 1986). Therefore, we have studied the effects of several selected nonionic polyethoxy derivatives of octylphenol (Triton X surfactant series) on sorption of 2-(1-naphthyl)acetic acid (NAA) for the isolated tomato fruit cuticle/buffer system.

We focused on sorption because it is an important component of membrane (cuticle) permeability (Nobel, 1974) and can be viewed as an early phase in the foliar absorption process. NAA is an important plant growth regulator (PGR) whose (a) general sorptive characteristics by plant cuticles have been investigated (Schönherr, 1976; Shafer and Bukovac, 1987a; Shafer et al., 1988) and (b) chemical characteristics are representative of weak organic acid type agrochemicals. A polyethoxy derivative of octylphenol (OP) with an average ethyleneoxy (EO) chain length of 9.5 was selected initially because it (a) is freely soluble in water, (b) has intermediate chemical characteristics in terms of octylphenoxy surfactants, and (c) is frequently used in agrochemical formulations.

EXPERIMENTAL SECTION

Plant Material/Cuticle Isolation. Locally field-grown mature tomato (*Lycopersicon esculentum* Mill. cv. Sprinter) fruit free of visual defects was selected for reasons previously discussed (Shafer and Schönherr, 1985). Disks, 20 mm in diameter, were punched from the fruit and incubated at 35 ± 1 °C in an aqueous mixture of pectinase (4%, w/v; ICN Nutritional Biochemicals), cellulase (0.4%, w/v; Sigma), and NaN_3 (1 mM) in 50 mM sodium citrate buffer at pH 4.0 (Orgell, 1955). NaN_3 was included to prevent bacterial and fungal growth. After 2 days and two changes of enzyme solution, the cuticle could be separated from the outer epidermal cell walls. Adhering cellular debris was removed with a jet of distilled water; the cuticles were air-dried and stored at 23 °C until used.

Isolated cuticles will be referred to as cuticular membranes (CM). CM extracted for 3 days with at least 10 changes of chloroform–methanol (1:1, v/v) at 50 °C to remove the epicuticular and cuticular waxes, i.e. soluble cuticular lipids (SCL), will be termed dewaxed cuticular membranes (DCM).

Radioisotope. Radioactive 2-(1-[1-^{14}C]naphthyl)acetic

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0021-8561/89/1437-0486$01.50/0

acid (sp act. 2.3 GBq mmol^{-1}; Amersham) with a purity of 98%, as determined by thin-layer radiochromatography, was used in this study.

Surfactant. 4-(1,1,3,3-Tetramethylbutyl)phenol with an EO side chain of 9.5 mol was used. The trade name (registered trademark, Rohm and Haas Co.) for this surfactant is Triton X-100. The CMC for OP + 9.5EO is 0.019% (w/v), and other selected properties of this surfactant relevant to foliar penetration were reported elsewhere (Stevens and Bukovac, 1987a).

This surfactant was representative of a commerical preparation. The EO number is an average value, with the ethoxymer mole ratio distribution following a Poisson distribution (Rothman, 1982; Triton Surface-Active Agents, 1982). No attempt was made to purify the surfactant preparation. All concentrations were based on weight/volume.

Measurement of Sorption. Sorption was measured for the CM/buffer and DCM/buffer systems by the procedure of Riederer and Schönherr (1984). Sodium citrate buffer (20 mM) at pH 3.2, containing 1 mM NaN_3, was used in all experiments.

Random samples (25–50) of CM or DCM disks were selected and sliced into small (approximately 1 mm × 10 mm) strips (preliminary results showed no significant effect of strip size). Weighed subsamples (approximately 5 mg) were placed into 5-mL glass vials, and 1.5 mL of ^{14}C-labeled NAA (300–550 nM) buffered solution was pipetted into each vial. Vials were closed with Teflon-lined screw caps and shaken horizontally in a water bath at 25 ± 0.5 °C.

At designated time intervals, 100-μL aliquots were removed, and radioactivity was determined by liquid scintillation spectrometry (LKB-Wallac LSC, Model 1211). Scintillation cocktail was composed of 1,4-dioxane (10 mL), containing 100 g of naphthalene and 5 g of diphenyloxazole (PPO) L^{-1}. All samples were counted to a 2σ error of approximately 1.0% and corrected for background. Since quenching was constant throughout the course of these experiments, all calculations were performed with CPM values. The amount of ^{14}C-labeled NAA sorbed by the CM or DCM was determined by subtracting the quantity of [^{14}C]NAA in the bulk dosing solution from the amount originally present (Kipling, 1965).

Apparent partition coefficients (K^{pH}) were calculated by

$$K^{pH} = \frac{^{14}\text{C label, cuticle phase (Bq kg}^{-1})}{^{14}\text{C label, aqueous phase (Bq kg}^{-1})} \quad (1)$$

Radioassay of solutions in control vials, containing only ^{14}C-labeled solution, indicated there was no significant change in ^{14}C-label concentration over the experimental periods. Therefore, the assumption that the decrease in ^{14}C label in the bulk solution represents that sorbed by, or associated with, the CM or DCM appears valid.

Spectrophotometric Measurement. Buffered (pH 3.2) solutions of NAA (100 μM), containing selected concentrations (percent, weight/volume) of a linear primary alcohol (C_{9-11}) polyethoxylate (10EO) surfactant (Neodol 91 Series, Shell Oil Co.), were used for a UV spectrophotometric assay (Gilford Instruments, Model 2600). The CMC for this surfactant (0.029%) was obtained from the literature (Lownds, 1987). OP + 9.5EO could not be used for this procedure since its UV absorption spectrum overlapped the NAA absorption spectrum [for OP + 9.5EO UV absorption spectrum, see Cardinal and Mukerjee (1978)].

Statistics. All measurements were made with five replications per treatment. For the time course measurements, the same five replicates were sampled repeatedly. The results are presented as the means with their coefficients of variation (Tables I and II) or 95% confidence intervals (Figures 1, 2, and 4–6).

Table I. Apparent Partition Coefficients (K^{pH}) for NAA and Tomato Fruit Cuticular Membranes (CM), As Affected by OP + 9.5EO, at Sorption Equilibrium (48 h), pH 3.2 and 25 °C

surfactant concn, % (w/v)	K^{pH}	surfactant concn, % (w/v)	K^{pH}
0	150.9 (3.8)[a]	4.0	0.2 (83.7)
1.0	20.8 (11.5)	8.0	0.0[b]
2.0	11.6 (21.6)	16.0	0.0[b]

[a] Mean of five replications with coefficient of variation in parentheses. [b] None detected.

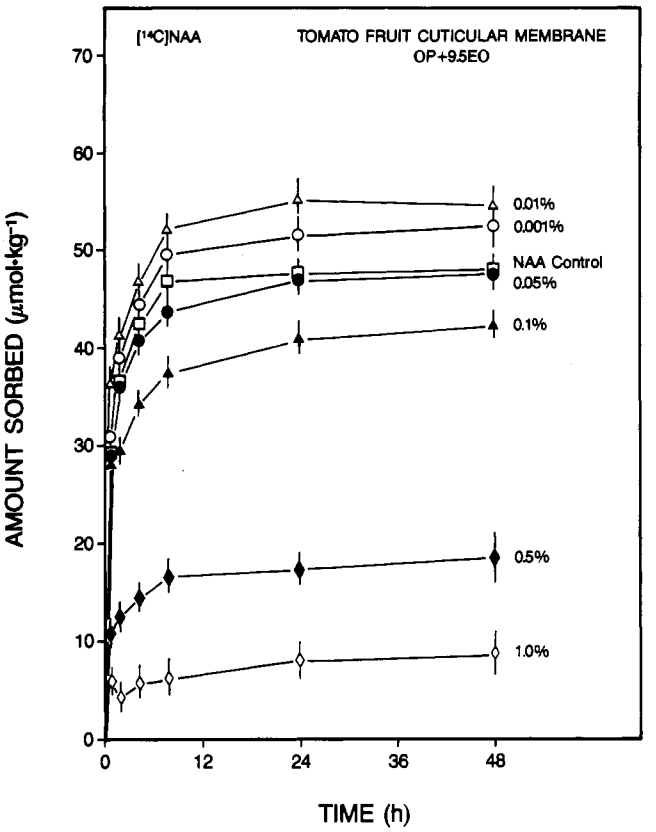

Figure 1. Effect of selected OP + 9.5EO concentrations (%, w/v) on time course of NAA sorption by tomato fruit cuticular membranes. Assay conditions: pH 3.2, 25 °C. Means of five replications and their respective confidence intervals (P = 0.05).

RESULTS AND DISCUSSION

Increasing surfactant concentration had a pronounced effect on sorption of NAA by both CM and DCM. For OP + 9.5EO concentrations below the CMC, there was little effect of the monomeric form on the time required for NAA to reach sorption equilibrium with CM (Figure 1) and DCM (data not shown) and on the equilibrium sorption values for NAA by CM and DCM (Figure 2). The equilibrium sorption values (micromoles per kilogram) for surfactant treatments were slightly higher than the control values. This was presumably due to enhanced wetting of CM or DCM because of lower surface tensions.

In contrast, for OP + 9.5EO concentrations above the CMC, there was an inverse relationship between sorption and concentration for both CM and DCM (Figures 1 and 2; Table I). At concentrations above 4%, no measurable amount of NAA was sorbed by CM (Table I). These data suggest that micelles depressed NAA sorption by CM and DCM.

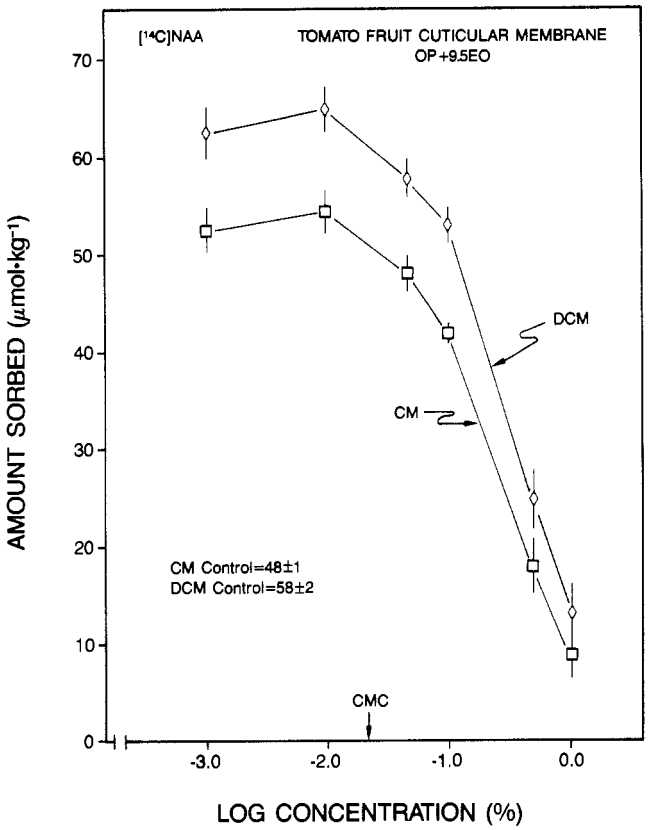

Figure 2. Effect of selected OP + 9.5EO concentrations (%, w/v) on sorption of NAA by tomato fruit cuticular membranes (CM) and dewaxed cuticular membranes (DCM). Assay conditions: 48 h, pH 3.2, 25 °C. Means of five replications and their respective confidence intervals (P = 0.05).

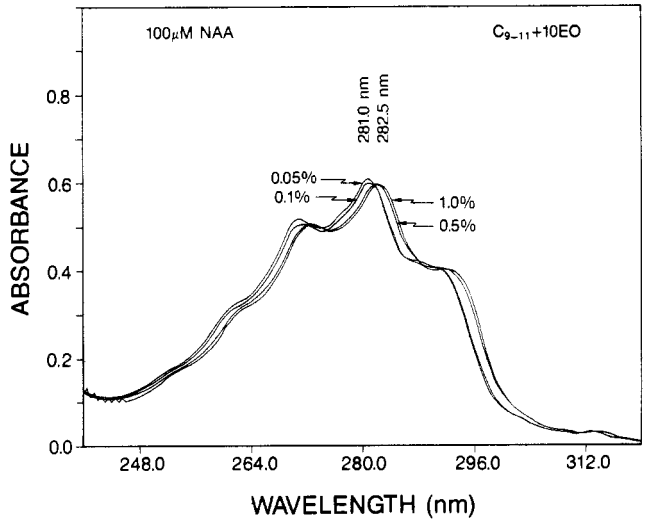

Figure 3. Absorption spectrum for 100 μM NAA as affected by selected linear primary alcohol ethoxylate (C_{9-11} + 10EO) concentrations. Assay conditions: pH 3.2, 25 °C.

The biphasic nature of OP + 9.5EO effects on NAA sorption by tomato fruit cuticles was qualitatively similar to the effects of surfactants on drug absorption [for a review, see Attwood and Florence (1983) and Florence and Gillan (1975a)]. Therefore, some insight into the mechanism of OP + 9.5EO action on depressing NAA sorption by tomato fruit cuticles may be obtained by examining pharmaceutical studies on drug/surfactant interactions.

Two possible mechanisms of surfactant action on decreasing drug absorption proposed by Florence and Gillan (1975b) may apply to our system, namely (a) physical blocking of NAA sorption sites by surfactant and/or (b) solubilization of NAA within surfactant micelles, thereby decreasing the bulk solution concentration of NAA

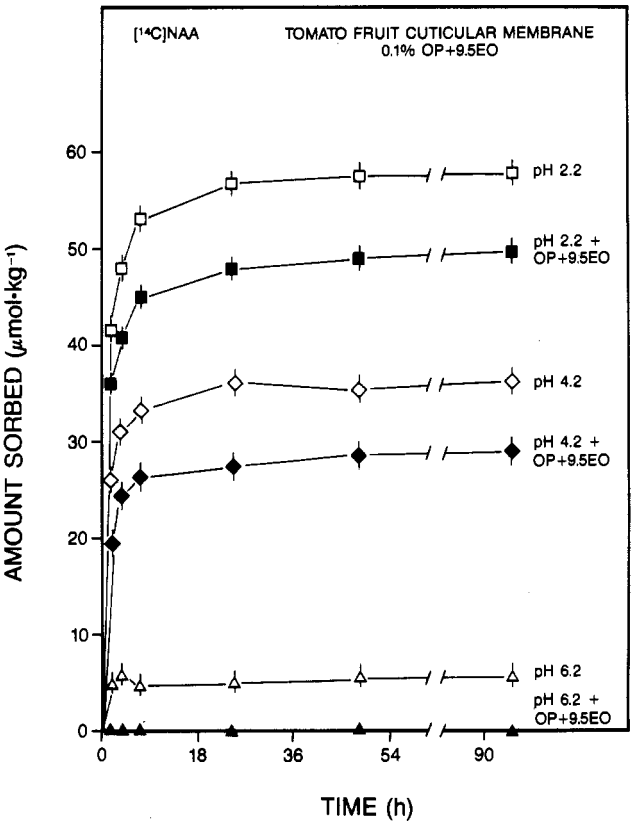

Figure 4. Effect of OP + 9.5EO (0.1%, w/v) on time course of NAA sorption by tomato fruit cuticular membranes at selected pH levels. Assay temperature: 25 °C. Means of five replications and their respective confidence intervals (P = 0.05).

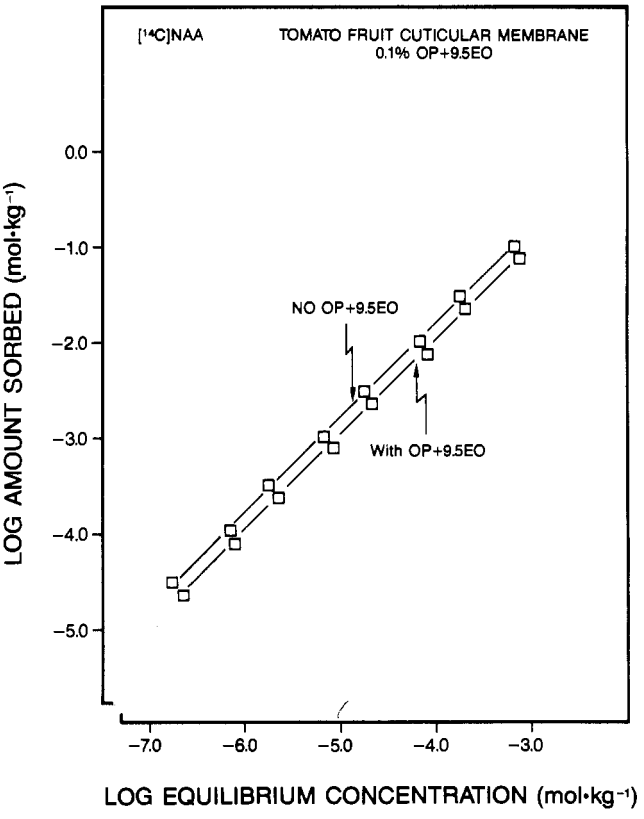

Figure 5. Effect of OP + 9.5EO (0.1%, w/v) on the sorption isotherm for NAA and tomato fruit cuticular membranes. Assay conditions: 48 h, pH 3.2, 25 °C. Means of five replications.

Table II. Apparent (K^{pH}) and pH-Corrected Partition Coefficients (K) for NAA and Tomato Fruit Cuticular Membranes, As Affected by OP + 9.5EO (0.1%, w/v), at Sorption Equilibrium (48 h) and 25 °C

pH	cuticular membrane	no surfactant		with surfactant		K(surf.)/K(no surf.) × 100
		K^{pH}	K^a	K^{pH}	K	
2.2	CM	170.2 (3.1)[b]	171.8 (3.6)	129.5 (4.9)	130.7 (4.8)	76.1
	DCM	204.4 (2.0)	206.3 (2.0)	161.6 (2.6)	163.1 (2.6)	79.1
3.2	CM	147.1 (3.5)	160.5 (3.5)	112.6 (2.0)	122.9 (2.0)	76.6
	DCM	193.5 (3.4)	211.1 (3.4)	150.8 (6.2)	164.5 (6.1)	77.9
4.2	CM	81.6 (3.2)	157.2 (2.3)	61.3 (3.3)	117.4 (3.4)	74.7
	DCM	91.6 (4.4)	176.0 (3.9)	81.3 (6.2)	155.5 (6.2)	88.4
6.2	CM	9.4 (12.8)	870.3 (13.7)	1.2 (58.3)	108.8 (58.5)	12.5
	DCM	11.6 (19.8)	1075.1 (20.4)	6.1 (104.9)	566.1 (103.9)	52.7

[a] K values calculated from eq 2, where pK for NAA was 4.24 (Dippy et al., 1954). [b] Mean of five replications with coefficient of variation in parentheses.

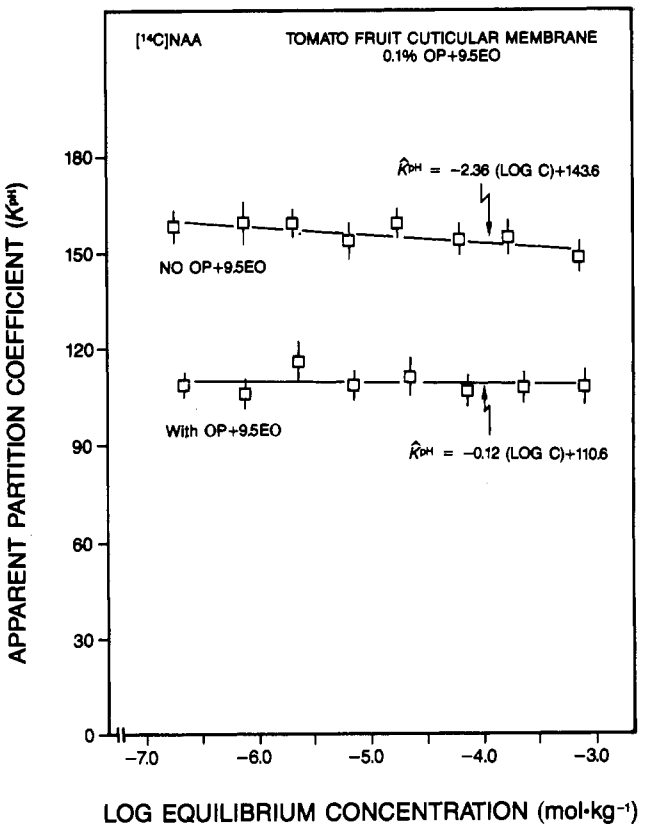

Figure 6. Effect of OP + 9.5EO (0.1%, w/v) on the concentration dependence of apparent partition coefficients (K^{pH}) for NAA and tomato fruit cuticular membranes. Assay conditions: 48 h, pH 3.2, 25 °C. Means of five replications and their respective confidence intervals (P = 0.05).

available for sorption. Either mechanism could account for decreased NAA sorption by CM or DCM.

The suggestion of physical blocking of NAA sorption sites by surfactant molecules is partially supported by OP + 9.5EO sorption data (Shafer and Bukovac, 1987b). Namely, below the CMC, sorption of OP + 9.5EO increased sharply as the CMC was approached. Immediately below and above the CMC, OP + 9.5EO sorption reached an apparent plateau. At surfactant concentrations in excess of the CMC (0.5 and 1.0%), sorption again increased sharply with increasing concentration. Data obtained with other members of the Triton X surfactant series also provide evidence that surfactants may become associated with the cuticle (Shafer and Bukovac, 1987b; Shafer et al., in press). The nature of the surfactant association with CM or DCM (e.g., adsorbed micelles, multilayer formation) has not been resolved.

There are some inconsistencies regarding our data and the physical blocking hypothesis however, and these deserve comment. Sorption of OP + 9.5EO, below the CMC, increased with increasing OP + 9.5EO concentration (Shafer and Bukovac, 1987b). However, NAA sorption by CM and DCM was not adversely affected by OP + 9.5EO until the CMC was exceeded. In fact, NAA sorption by CM and DCM increased slightly at pre-CMC concentrations (Figure 2). This suggests that NAA sorption sites for OP + 9.5EO and NAA were not the same, at least at pre-CMC OP + 9.5EO concentrations. Another concern for the physical blocking hypothesis deals with OP + 5EO, a polyethoxy derivative of octylphenol with 5EO (Triton X-45; registered trademark, Rohm and Haas Co.). Sorption of OP + 5EO by tomato fruit CM was independent of OP + 5EO concentration from 0.5 to 1.0% (Shafer and Bukovac, 1987b). However, the amount of NAA sorbed by tomato fruit CM decreased from 65 to 23 μmol·kg^{-1} when the OP + 5EO concentration was increased from 0.5 to 1.0% (Shafer and Bukovac, 1988). This suggests, at least for OP + 5EO and NAA, that physical blocking of NAA sorption sites by OP + 5EO molecules was not responsible for reduced sorption of NAA by tomato fruit CM.

With respect to the possible solubilization of NAA by OP + 9.5EO micelles, there are numerous reports demonstrating that drugs (active ingredients) may become associated with surfactant micelles [for a review, see Attwood and Florence (1983)]. In aqueous systems, surfactant-enhanced solubilization of an insoluble solute is generally negligible until the CMC is reached or exceeded (Goodhart and Martin, 1962; Rosen, 1978). Solubilization is clearly a micellar phenomenon (Attwood and Florence, 1983; Florence and Gillan, 1975a; Gibaldi and Feldman, 1970; Mukerjee, 1980). While NAA was soluble in our buffered solutions in the absence of OP + 9.5EO (NAA solubility in water at 20 °C, 420 mg·kg^{-1}; Worthing, 1983), the association of relatively polar solutes with surfactant micelles has been observed (Gouda et al., 1970). The fact that NAA sorption by CM and DCM did not decrease until the CMC of OP + 9.5EO was exceeded (Figure 2) supports the suggestion that NAA was solubilized by, or partitioned into, OP + 9.5EO micelles. This would lead to a lower bulk solution concentration of NAA and consequently lower NAA sorption by CM and DCM.

Evidence for NAA/micellar association in our system was obtained spectrophotometrically on buffered solutions of NAA and a linear primary alcohol polyethoxylate surfactant (C_{9-11} + 10EO) (Figure 3). The non-UV-absorbing linear primary alcohol polyethoxylate used was considered sufficiently similar chemically to OP + 9.5EO in terms of hydrophobe/hydrophile chain lengths to serve as a model surfactant. The magnitude of change in the wavelength of maximum absorption for NAA in the presence of the C_{9-11} + 10EO surfactant was small, from 281.0 (NAA in buffer only) to 282.5 nm, compared to the change seen in studies examining surfactant effects on anthraquinoid acid

dyes (Tuong and Hyano, 1977; Tuong et al., 1977). However, the shift in the absorption maximum for NAA occurred only when the C_{9-11} + 10EO concentration was 0.5% or greater. At 10% surfactant concentration, maximum absorption was still at 282.5 nm (data not shown). The most likely reason for the spectral shift at concentrations greater than the CMC (0.029%) was the NAA solubilization capacity of the micelles. At 100 μM NAA, a given threshold concentration of C_{9-11} + 10EO was apparently required before the NAA was completely solubilized by, or became associated with, the micelles. The spectral shift was presumably due to micellar microenvironmental polarity effects on the NAA molecule (Mukerjee and Cardinal, 1978).

It is clear that OP + 9.5EO at post-CMC concentrations dramatically decreased NAA sorption by CM and DCM. However, the question of whether OP + 9.5EO micelles physically block NAA sorption sites or whether OP + 9.5EO micelles solubilize NAA, thereby decreasing the bulk solution concentration of NAA available for sorption, cannot be completely resolved at this time. While there are data supporting both processes, it appears that solubilization of NAA by OP + 9.5EO micelles was of greater significance in our system. It may well be that both mechanisms were involved.

Sorption of NAA by CM (Figure 4) and DCM (at sorption equilibrium; Table II) was dependent on solution pH. OP + 9.5EO (0.1%) lowered NAA sorption at each pH level. Solution pH was of critical importance since both degree of NAA dissociation (pK 4.24; Dippy et al., 1954) and net cuticular charge (p$I \approx 3.0$; Schönherr and Huber, 1977; Shafer, 1984) were dependent on H ion concentration. K^{pH} values reflect the amount of NAA sorbed by CM or DCM at equilibrium, without correction for degree of NAA dissociation. In our study, K^{pH} values obtained at a given solution pH may be compared with one another. However, for comparison of NAA sorption at different pH levels, corrections for NAA dissociation must be made.

If the assumption is made that only the nondissociated species is sorbed by the cuticle, K^{pH} values can be corrected for pH effects by (Riederer and Schönherr, 1984)

$$K = K^{pH}(1 + 10^{pH-pK}) \quad (2)$$

giving the partition coefficient (K). The K values for NAA sorption by CM and DCM, with and without OP + 9.5EO (0.1%), are summarized in Table II. OP + 9.5EO micelles consistently lowered K values for both CM and DCM.

There is some evidence (Schönherr, 1976; Riederer and Schönherr, 1984) that only the nondissociated NAA molecule is sorbed by the plant cuticle. However, other data suggest that some dissociated ions of weak organic acids may penetrate the cuticle (Greene and Bukovac, 1972). If sorption of the anionic species occurs, the calculated K values obtained with eq 2 at pH levels above the pK would be in error. Our K values obtained for NAA with CM and DCM at values pH 2.2–4.2 were in relatively good agreement with one another (Table II). The calculated K values using the K^{pH} values obtained at pH 6.2, however, were approximately 4–5 times greater than the K values obtained at the lower pH levels. These values also had large coefficients of variation. Schönherr (1976) has attributed the small amount of sorption at pH levels above the pK to the presence of a lipophilic radiochemical contaminant whose sorption becomes dominant at higher pH levels. An alternative possibility is that there are domains in the cuticle that may sorb polar compounds (Franke, 1967; Schönherr and Bukovac, 1973). Nevertheless, the degree of sorption above the pK is limited (Table II; Figure 4).

The amount (mole per kilogram) of NAA sorbed by CM increased with increasing NAA concentration (Figure 5). In a previous study (Shafer et al., 1988), we determined that sorption of NAA by tomato cuticles was in the form of the type C, or constant partitioning, isotherm (Giles et al., 1960). This isotherm form is characteristic of when the sorbate (NAA) penetrates the sorbent (cuticle) more easily than the solvent (water) and of a sorbent with flexible molecules and differing degrees of crystallinity. In the presence of OP + 9.5EO (0.1%), the amount of NAA sorbed was less for all NAA concentrations examined but the shape of the isotherm was not affected. This effect of OP + 9.5EO on NAA sorption by CM is more readily apparent from K^{pH} values (Figure 6). K^{pH} values for NAA and CM decreased slightly with increasing NAA concentration, a response pattern previously observed with tomato fruit cuticles and 2,4-D (Riederer and Schönherr, 1984). OP + 9.5EO (0.1%) significantly lowered K^{pH} values for NAA across the entire NAA concentration range examined. While these data do not provide direct evidence of NAA solubilization by OP + 9.5EO micelles, it is interesting to note that previous reports have shown that partition ratios for iodoform and triiodophenol between micelles and the bulk solution were independent of drug concentration [as reviewed by Gibaldi and Feldman (1970)].

On the basis of our results, three areas deserve further study. First, given the relationship between sorption and membrane (cuticle) permeability (Nobel, 1974), it would be interesting to determine whether OP + 9.5EO micelles decrease NAA penetration across isolated plant cuticles. Second, the question arises whether the general relationships provided by these results are useful in predicting foliar absorption. Some data generated in our laboratory with NAA-induced ethylene evolution suggest that surfactant concentrations in excess of the CMC reduce NAA penetration into plant foliage (Lownds and Bukovac, 1983; Shafer and Bukovac, unpublished data). Third, similar data are needed on other agriculturally important surfactants.

ACKNOWLEDGMENT

We thank A. Y. S. Yang, E. I. du Pont de Nemours & Co., Wilmington, DE, for stimulating discussions.

Registry No. NAA, 86-87-3; Triton X-100, 9002-93-1.

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Received for review February 28, 1988. Accepted August 22, 1988.

Michigan Agriculture Experiment Station Journal Article No. 12567. This paper is based on work supported, in part, by U.S. Department of Agriculture—Agricultural Research Service Grants CWU 3607-20300-004-01 S and 58-5114-7-1002 and by a grant from the Shell Development Co., Modesto, CA 95352. Portions of this work were previously given in poster presentations (Shafer and Bukovac, 1985; Shafer et al., 1987).

Fate of the Fungicide Furalaxyl in the Nutrient Solution of Tomato Crops by the Nutrient Film Technique

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The fungicide furalaxyl [methyl *N*-(2,6-dimethylphenyl)-*N*-(2-furanylcarbonyl)-DL-alaninate] was incorporated into the nutrient solution of tomato crop grown according to the nutrient film technique (NFT). Incorporation into the refreshed nutrient solution was made several times at different plant growth stages. Furalaxyl was decomposed in the nutrient solution into *N*-(2,6-dimethylphenyl)-*N*-(2-furanylcarbonyl)-DL-alanine (**2**) and *N*-(2,6-dimethylphenyl)-DL-alanine (**4**) by enzymatic processes. Half-lives in the nutrient solution of furalaxyl and of the total furalaxyl plus compounds **2** and **4** at the beginning of the crop were 2.5 and 5.2 days, respectively. With starting of the continuous recirculation of the nutrient solution, they increased to 6 and 12 days; with plant development, they decreased progressively to 3.5 and 6.5 days. No furalaxyl nor compounds **2** or **4** were detected in tomato fruit, the limit of sensitivity being 0.02 ppm (of fresh weight).

Present-day horticultural production of tomatoes in Northern Europe is now based almost entirely on artificial substrates rather than soil, the common practice until 15 years ago, basically due to the cost of sterilization of the soil due to the large increase in the cost of oil and the lack of good soil and the cost of resoiling houses when the soil is exhausted (Wilson, 1986). Artificial substrates have the following advantages: They are disease and weed free and are light in weight. Repetitive mixes have the same composition. They show quicker growth and higher yields. Tomato yields have increased in the last 30 years mainly due to monocropping systems and growing out of the soil (Wilson, 1983). However, plant protection problems begin to arise now in soilless culture of tomato, especially from the fungi *Pythium* and *Phytophthora nicotianae* (Vanachter et al., 1986). One or two fungicide treatments are made during one crop, in commercial practice. One treatment is always made at the beginning of the crop, when the damage made to the plant roots during plantation makes the plants very susceptible to the fungi. The second treatment is made when symptoms of sickness arise (Benoit and Ceustermans, 1987). Furalaxyl is a fungicide widely used in the soilless culture of tomato. It gives very good plant protection and does not give residues in the fruits at harvest.

The residual behavior of furalaxyl has been studied in greenhouse tomatoes grown on soil sprayed with furalaxyl (Cabras et al., 1985). To our knowledge, nothing has been published about the uptake and translocation of furalaxyl by plants and about its metabolism in biological systems. However, the uptake and translocation of the fungicide metalaxyl has been studied in sunflower plants (Marucchini et al., 1983). Metalaxyl has the chemical structure of furalaxyl in which the 2-furanylcarbonyl group has been replaced by 1-methoxyacetyl. Furalaxyl thus should also be systematic. The metabolism of metalaxyl has been studied in lettuce and sunflower (Businelli et al., 1984).

In the present work, we studied the metabolism of furalaxyl in the nutrient solution of tomato NFT crop grown on a semicommercial scale.

EXPERIMENTAL SECTION

Tomato Crop with the Nutrient Film Technique (NFT): Furalaxyl Treatments. Tomato plants were grown in semicommercial installations. Seeds of tomato cv. Concreto were sown in peat–sand on 11-21-86. Seedlings were transferred to cubes of rockwool (10 × 10 × 17 cm) on 12-10-86 and placed in the NFT gullies on 1-28-87 at the Proefstation voor de Groenteteelt, St Katelijne-Waver, Belgium, when the buds of the first truss were just visible. Plant interdistance in the same row was 60 cm; interdistance between each row was 1 m; there were six rows in a glasshouse that was 6.5 m wide. During the propagation stage (until 2-16-1987), the Cooper nutrient solution (Cooper, 1979) was circulated for five 15-min periods every 24 h (four periods during daylight hours and one period during darkness). Subsequently, the nutrient solution was continuously recirculated. The nutrient solution was maintained at pH 5.8. The conductivity of the nutrient solution was continuously monitored by two electrodes and was regulated by separate addition of water and of a concentrate solution of the Cooper mixture; this last was made up instantaneously by pumping from two separate tanks, the one containing the calcium nitrate solution and the other containing the solution of the other salts. The pH of the nutrient solution was also continuously monitored by two electrodes and was adjusted by pumping from two tanks of acid and basic solutions. The

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0021-8561/89/1437-0492$01.50/0