# An Automated Aqueous Stability Study with the Experimental Insecticide Nifluridide

At 25 °C, the experimental insecticide nifluridide [*N*-[2-amino-3-nitro-5-(trifluoromethyl)phenyl]-2,2,3,3-tetrafluoropropanamide, coded EL-468] rapidly dissipated from water at three different pH levels. The rate of reaction proceeded via first-order kinetics with half-lives of 15.5, 3.5, and 2.0 h at pH 5.0, 7.0, and 9.0, respectively. The disappearance of nifluridide was accompanied by a corresponding formation of a cyclized product, 7-nitro-2-(1,1,2,2-tetrafluoroethyl)-5-(trifluoromethyl)benzimidazole (coded EL-919). The rate study was automated by programming a Waters Intelligent Sample Processor (WISP) to inject the reaction solutions directly into a reverse-phase high-pressure liquid chromatographic system at specified time intervals. Nifluridide and its cyclized product were separated and measured on the same chromatogram. Detection was accomplished with a UV absorbance detector operated at a fixed wavelength (254 nm).

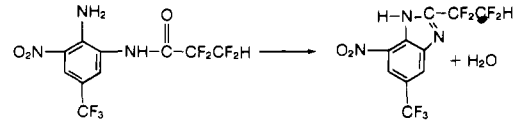
Nifluridide [*N*-[2-amino-3-nitro-5-(trifluoromethyl)-phenyl]-2,2,3,3-tetrafluoropropanamide, coded EL-468] is an experimental insecticide that is being evaluated for the control of the imported fire ant (*Solenopsis germinata*) and the red imported fire ant (*Solenopsis invicta*) in the southern United States. Broadcast applications of formulated ant baits containing 0.75% nifluridide at rates of 10–20 g of active ingredient/ha have resulted in delayed toxicity to ants, which permits distribution of the material throughout the colonies (Lilly Research Laboratories, 1981). It has been postulated that the delayed toxicity of nifluridide is due to its conversion to another compound with enhanced insecticidal properties when ingested by ants (Day et al., 1982). An aqueous stability study was conducted to help predict the persistence and environmental fate of the insecticide, and the results are reported here.

## EXPERIMENTAL SECTION

**Preparation of Solutions.** Buffered solutions were prepared at pH 5.0, 7.0, and 9.0 in HPLC-grade water from solutions of sodium acetate–acetic acid, boric acid–sodium hydroxide, and sodium bicarbonate–sodium carbonate, respectively. The reaction solutions were prepared with nifluridide at a concentration of 25 μg/mL in each buffered solution. Analytical standard solutions were prepared at 20 μg/mL in HPLC-grade acetonitrile for nifluridide and in 55:45 methanol–water (both HPLC grade) for the cyclized product, EL-919 [7-nitro-2-(1,1,2,2-tetrafluoroethyl)-5-(trifluoromethyl)benzimidazole]. The latter two solutions served as direct standards for determining the rate of dissipation of nifluridide in the buffered solutions.

**Analysis of Solutions.** Analysis of the reaction solutions was accomplished by injecting 40 μL directly into a reverse-phase high-pressure liquid chromatographic (HPLC) system. The HPLC system consisted of a Co-Pell ODS guard column (Whatman, Inc.), a μBondapak $C_{18}$ column (Waters Associates), a Waters Model 6000A solvent delivery system with methanol–water (55:45) at 1.0 mL/min, a Waters Model 440 UV absorbance detector (254 nm), and a Waters Model 710 Intelligent Sample Processor (WISP). The hydrolysis and direct standard solutions were injected into the HPLC system at predetermined intervals, which were programmed into the WISP (see Figures 2–4).

The concentrations of nifluridide and EL-919 were determined by comparison of the peak height responses of the compounds in the aqueous solutions with the peak heights in the direct standards. A measure of material




NIFLURIDIDE EL—919

**Figure 1.** Conversion of nifluridide to EL-919.

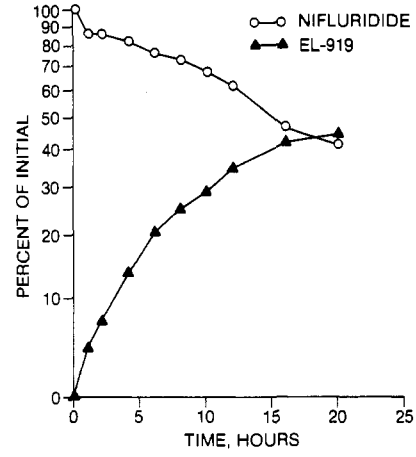



**Figure 2.** Rate of reaction of nifluridide at pH 5.0 with the formation of EL-919.

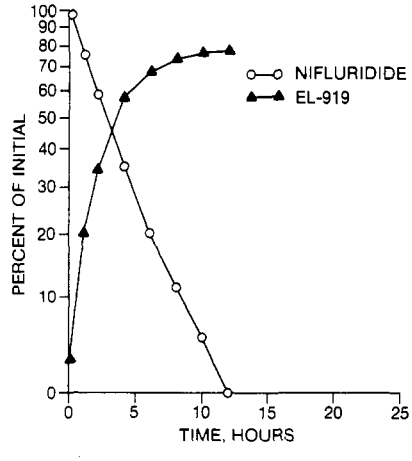



**Figure 3.** Rate of reaction of nifluridide at pH 7.0 with the formation of EL-919.

0021-8561/82/1430-0795$01.25/0 

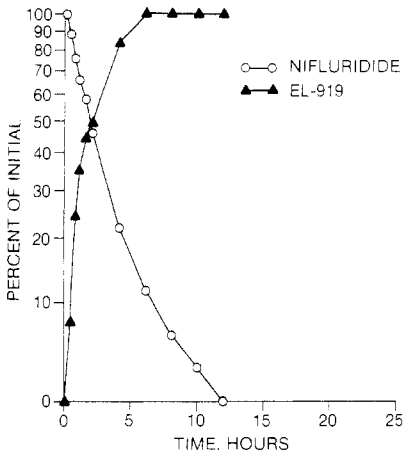



**Figure 4.** Rate of reaction of nifluridide at pH 9.0 with the formation of EL-919.

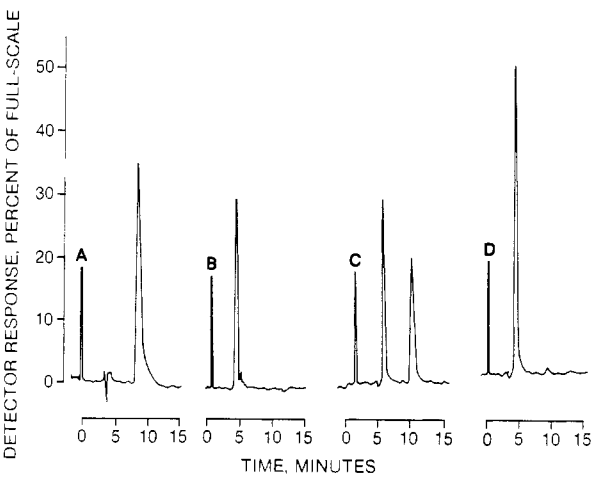



**Figure 5.** Chromatograms demonstrating the dissipation of nifluridide and the formation of EL-919 at pH 9.0: (A) nifluridide standard, 0.8 μg; (B) EL-919 standard, 0.8 μg; (C) reaction solution at 2 h (45.8% nifluridide, 49.1% EL-919); (D) reaction solution at 10 h (2.9% nifluridide, 105.7% EL-919).

balance was obtained by adding together the concentrations of nifluridide and EL-919 (as its nifluridide molecular weight equivalent).

## RESULTS AND DISCUSSION

The rapid conversion of nifluridide to EL-919 proceeds as indicated in Figure 1. The rates of disappearance and the corresponding formation of EL-919 are plotted on a semilogarithmic scale in Figures 2–4. The cyclization proceeded via a first-order reaction with half-lives of 15.5, 3.5, and 2.0 h for EL-468 at pH 5.0, 7.0, and 9.0, respectively. The total amount of nifluridide and EL-919 present in the final samples analyzed provided a good material balance (80–100%) compared to the initial amount of chemical in the aqueous solutions (Figures 2–4). No other hydrolysis products were observed.

On the basis of these results, nifluridide would likely dissipate very rapidly in natural water or in the presence of soil moisture. Thus, the parent compound would not be expected to persist in the environment.

Chromatograms demonstrating the determination of both compounds in the aqueous solutions are shown in Figure 5. The direct injection of the solutions into the reverse-phase HPLC system at specified intervals, which were programmed into the WISP, provided an automated rate study by eliminating the need to extract the buffered solutions with a suitable solvent, concentrate the sample extract, and redissolve the residue in a suitable solvent for injection into the HPLC. It is probable that this direct injection technique could also be applied to many other compounds of environmental interest that are highly labile in aqueous solution.

## LITERATURE CITED

Day, E. W.; Dorulla, G. K.; West, S. D.; Decker, O. D., presented at the 183rd National Meeting of the American Chemical Society, Las Vegas, NV, March 1982.

Lilly Research Laboratories "Technical Report on EL-468 Experimental Insecticide"; Lilly Research Laboratories: Indianapolis, IN, 1981.

**Sheldon D. West***
**Edgar W. Day, Jr.**

Agricultural Analytical Chemistry
Lilly Research Laboratories
Division of Eli Lilly and Company
Greenfield, Indiana 46140

Received for review December 18, 1981. Accepted April 23, 1982.

# Reinvestigation of the Alkaloids of *Lupinus sericeus* Pursh. Identification of a New Natural Product, 10,17-Dioxo-β-isosparteine

In addition to two previously reported *Lupinus sericeus* constituents, sparteine and β-isosparteine, 10,17-dioxo-β-isosparteine, a new natural product, and 17-oxosparteine, 10-oxo-β-isosparteine, and nuttalline (4β-hydroxylupanine) were also found in the aerial parts of this toxic range plant. Anagyrine, a suspected teratogenic constituent of this lupine for cattle, was not detected in the sample of this species under investigation. (−)-Sparteine exhibited the most potent acute toxic response for mice among the *L. sericeus* alkaloidal constituents tested.

*Lupinus sericeus* Pursh (silky lupine), a range plant indigenous to the Rocky Mountain states, has been cited as being one of the six most toxic lupines for sheep in the United States and Canada (Kingsbury, 1964; Clarke, 1970).

0021-8561/82/1430-0796$01.25/0 