

Table II. Recovery Studies^a

Herbicides	Potatoes			Carrots			Oranges, Peas, Corn,	Pine-apple,	Aspar-agus	Spinach			
	1.0	0.1	0.01	1.0	0.1	0.01				0.1	0.1	0.1	0.01
Linuron	99	97	100	92	98	96	100	98	98	93	98	102	100
Monuron	65	75	93	—	65	84	70	62	—	58	64	76	85
Diuron	81	84	95	—	78	98	84	87	74	68	80	95	100

^a Percent recovery at indicated spiked values (in parts per million). Each value is the average of duplicate analyses. Dashes indicate that no analyses were performed.

evaporating the final extract to dryness to remove any CH₃I present and then redissolved in hexane. This treatment resulted in a lower background in the chloride mode. Other columns used to confirm in the chloride mode were 6% DC 200, 5% SP 525, and 5% OV 17. All of these proved satisfactory for direct confirmation at the 0.1-ppm level.

Bleeding of the liquid phases was found to cause some problems in the chloride mode. SE 30/QF1 gave a consistently high background and could not be used at the 1× attenuation. OV 17 gave the least interference. In general, any halogen-containing liquid phase should be avoided in the chloride mode. In the nitrogen mode no interferences were observed with SE 30/QF1 although QF1 contains fluorine. Use of this phase necessitated routine (once a month) replacement of the acid gas scrubber at the end of the pyrolysis tube.

CONCLUSION

The developed method was found promising for the routine screening, quantitation, and confirmation of most *N*-phenylcarbamate and *N*-phenylurea herbicides by GLC

with electrolytic conductivity detection. Several hundred crop samples were passed through the GLC system without contamination of the pyrolysis tube or chromatography column.

LITERATURE CITED

- Buser, H., Grolimund, K., *J. Assoc. Off. Anal. Chem.* **57**, 1294 (1974).
 Cochrane, W. P., Greenhalgh, R., 166th National Meeting of the American Chemical Society, Chicago, Ill., Aug 1973.
 Fishbein, L., Zielinski, W. L., *J. Chromatogr.* **20**, 9 (1965).
 Gutenmann, W. H., Lisk, D. J., *J. Gas Chromatogr.* **4**, 424 (1966).
 Harris, R. J., Whiteoak, R. J., *Analyst* **97**, 294 (1972).
 Katz, S. E., Strusz, R. F., *J. Agric. Food Chem.* **17**, 1409 (1969).
 Kirkland, J. J., *Anal. Chem.* **34**, 428 (1972).
 Lawrence, J. F., *J. Agric. Food Chem.* **22**, 936 (1974).
 Lawrence, J. F., Iverson, F., *J. Chromatogr.* **103**, 341 (1975).
 Lawrence, J. F., Sen, N. P., *Anal. Chem.* **47**, 367 (1975).
 Lisk, D. J., *J. Agric. Food Chem.* **12**, 46 (1964).
 McKone, C. E., Hance, R. J., *J. Chromatogr.* **36**, 234 (1968).
 Onley, J. H., Yip, G., *J. Assoc. Off. Anal. Chem.* **54**, 1366 (1971).
 Spengler, D., Hamroll, B., *J. Chromatogr.* **49**, 205 (1970).
 Tanaka, F. S., Wien, R. G., *J. Chromatogr.* **87**, 85 (1973).
 Thier, H. P., *Angew. Chem.* **10**, 860 (1971).

Received for review May 13, 1975. Accepted July 24, 1975.

Photosensitization and Luminescence of Picloram

Bobby L. Glass

The photosensitization of picloram (4-amino-3,5,6-trichloropicolinic acid) was studied with known triplet-energy sensitizers. Benzophenone and benzoate increased the rate of photodecomposition of picloram by factors of about 3 and 2, respectively, upon photolysis (313 nm) in aqueous solution (pH 9.3). The quantum yields for the disappearance of picloram (aerated) with 254- and 313-nm light were 0.037 and 0.04, respectively. Phosphorescence emission of picloram in EPA-

glass at 77 K occurred at 450 nm with a radiative lifetime of 40 msec for the free acid and 32 msec for the methyl ester, thus indicating an n,π^* triplet state. Fluorescence emission was quenched in weakly acidic, neutral, and basic solutions, but emitted strongly at 425 nm in aqueous sulfuric acid solutions at pH <1.16. The conclusion drawn from these results is that photodecomposition of picloram takes place from the excited triplet state.

Applications of picloram (4-amino-3,5,6-trichloropicolinic acid) to Texas' rangelands and forests have been effective in the control of a variety of undesirable woody plants and herbaceous weeds (Bovey and Scifres, 1971). Sensitive crops, such as soybeans, suffer detrimental effects from picloram in soils and residues transported by runoff water from treated areas. Picloram has been shown to photodecompose in aqueous solution in the laboratory by direct

photolysis with 254 nm (Hall et al., 1968) and with ~366 nm (Mosier and Guenzi, 1973). The dechlorination of picloram appears to be the most clearly defined photochemical reaction that results from the photolysis of picloram. Hall et al. demonstrated and Mosier and Guenzi later confirmed that two chloride ions are liberated for each molecule of picloram that undergoes photodecomposition. Very little success has been obtained in isolating and identifying photoproducts of picloram, thus limiting the mechanistic characterization of the degradation process. A better knowledge of its photochemical degradation is necessary to fully assess the fate of picloram and its photoproducts in the aquatic environment.

Agricultural Chemicals Management Laboratory, AEQI, Agricultural Research Service, U.S. Department of Agriculture, Beltsville, Maryland 20705.

The objective here was to determine if the photosensitization of picloram occurred with triplet energy sensitizers. The quantum yields for the disappearance of picloram and the percent yields of $^{14}\text{CO}_2$ liberated by decarboxylation were determined with 313- and 254-nm light. The fluorescence and phosphorescence spectra of picloram were measured in order to determine the nature of the excited states that arise from photolysis.

EXPERIMENTAL SECTION

Reagents and Solutions. Analytical grade picloram (99.9% purity) and ^{14}C -carboxyl labeled picloram (4.13 $\mu\text{Ci}/\text{mg}$) were supplied by Dow Chemical Co. The sensitizers, benzophenone, benzoic acid (anion), acetone, phenol, and benzene, were commercially obtained and used without further purification. In preparing a stock picloram solution, 0.25 g of inactive picloram and 40 ml of a ^{14}C -labeled picloram solution ($1.3 \times 10^{-5} M$) were mixed with 250 ml of a 0.025 M sodium borate solution. This buffered solution of picloram was adjusted to pH 9.3 with 0.01 N sodium hydroxide and diluted to 1 l. with distilled water. Aliquots of the $1.1 \times 10^{-3} M$ picloram solution were used with and without sensitizers throughout the experiment.

Quantum Yields. Measurements of the quantum yields for the disappearance of picloram in the presence of benzophenone and benzoate were determined separately in photolysis with 254- and 313-nm light. Excitation with 254-nm light was conducted with a 5.6-W PCQ-XI photochemical reactor (Ultraviolet Products Inc.) that consisted of four 7.5 cm (i.d.) circular low-pressure mercury lamps, enclosed with an aluminum reflector. Light intensity measurements were made with the potassium ferrioxalate actinometer as described by Hatchard and Parker (1956). Cylindrical vycor cells (200 ml, 26×3.5 cm) were used to photolyze 100-ml aliquots of the picloram solution ($1.1 \times 10^{-3} M$). Deaerated solutions were prepared by bubbling through dry nitrogen for 2.5 hr. Photolysis generally was performed for 0.5 hr at a temperature of $32 \pm 2^\circ\text{C}$.

Photolysis with light at 313 nm was conducted with a Hanovia 450-W medium-pressure mercury arc, filtered through a Pyrex sleeve ($\lambda > 290$ nm). A potassium chromate solution ($5 \times 10^{-4} M$), which was circulated through the outer jacket (~ 1.5 -cm layer) of the immersion well, acted simultaneously as a coolant and a chemical filter. The chromate solution transmitted 70% of the 313-nm mercury line, but was opaque to the 366-nm line and to all light < 290 nm. The 12-l. volume of chromate solution was renewed after each experiment, as recommended by Hunt and Davis (1947). Aliquots (250 ml) of the picloram solution with and without sensitizers were photolyzed for 4 hr at $31 \pm 1^\circ\text{C}$.

The unreacted picloram was analyzed on a F&M Scientific Model 700 gas chromatograph (E.C. Detector). A 4-ft glass column (120 cm \times 3 mm) packed with 3% OV-3 on 60–80 mesh Chromosorb W-HP was maintained at 200°C . An argon–methane (95%:5%) mixture was used as the carrier gas with a flow rate of 60 cm^3/min . Aliquots (5 ml) of the photolyzed picloram solutions were added to 100 ml of water (pH 1.0). The free acid of picloram was extracted into 100 ml of ethyl ether and dried over anhydrous sodium sulfate. The methyl ester of picloram was prepared with ethereal diazomethane and quantitated by comparing peak areas with those of known quantities of the standard compound.

Percent Yields of $^{14}\text{CO}_2$. Twenty-five-milliliter aliquots were removed from the sensitized and unsensitized picloram solutions after photolysis and added to 15 ml of inactive sodium carbonate solution ($1 \times 10^{-3} M$). The mixture was acidified with 10 ml of 2% (v/v) HNO_3 and 200 cm^3 of nitrogen was bubbled through at 10 cm^3/min . The liberated carbon dioxide was first bubbled through 50 ml of dilute HNO_3 with a gas scrubber that had a fritted end. The $^{14}\text{CO}_2$ was then trapped in a second gas scrubber that con-

tained 50 ml of 0.5 N NaOH. Aliquots (1 ml) of the alkaline solution were mixed with 20 ml of liquid scintillant containing 4 g of PPO (2,5-diphenyloxazole), 0.2 g of POPOP (1,4-bis[2-(5-phenyloxazolyl)]benzene), 60 g of naphthalene, 100 ml of methanol, and 20 ml of ethylene glycol, which were diluted to 1 l. with dioxane. The radioactivity was measured with a Packard scintillation counter. The accuracy of the $^{14}\text{CO}_2$ measurement was not determined in the study; however, in a similar decarboxylation investigation with benzoate, Matthews and Sangster (1965) reported an error of $\pm 8\%$ for the method.

Emission Spectra. Fluorescence and phosphorescence emission spectra of picloram were obtained on an Aminco-Bowman spectrophotofluorometer equipped with a 250-W xenon arc lamp and a photomultiplier tube (1P21). All the fluorescence measurements were made in a conventional quartz fluorescence cell at room temperature. Aqueous solutions of picloram with varying pH values were used in the fluorescence measurements. The pH values were adjusted with aqueous solutions of sodium hydroxide or sulfuric acid, which were both ACS reagent grade. All measurements were made on picloram solutions with final concentrations of $10^{-3} M$. A rotating can phosphoroscope was used for measuring the phosphorescence emissions. The phosphorimetry solvent used was EPA (ethyl ether–isopentane–ethanol, 5:5:2, v/v). Ethyl ether (Burdick and Jackson Lab, Inc.), isopentane (Eastman Kodak Co.), and absolute ethanol (U.S.I.) were all spectroscopic grade and used without further purification. Aliquots of the two forms of picloram (free acid and ester) dissolved in EPA were placed in separate quartz capillary sample tubes. They were individually inserted into a quartz window dewar flask that contained liquid nitrogen. Both the uncorrected fluorescence and phosphorescence spectra were obtained on a strip-chart potentiometric recorder.

Phosphorescence lifetime measurements were determined on a Phosphor Standard Calibrator (Greenough and Hammond, 1974), which was designed for the purpose of standardizing phosphorescent and fluorescent activity of the luminescent coating applied to postage stamps (U.S. Postal Service). The instrument consisted of a pulse uv lamp (254 nm), photomultiplier tube (9558 QMA), an oscilloscope, and signal amplification and readout components that included a binary counter. As a reference sample, benzophenone gave a phosphorescence lifetime of 6 msec in EPA at 77 K. This measurement agreed well with the 7-msec lifetime, determined with a similar pulse source phosphorimeter (Harbaugh et al., 1973).

RESULTS AND DISCUSSION

Photosensitization. The percentages of picloram that underwent photodecomposition in the presence of known triplet-energy sensitizers are shown in Table I. These data were obtained from sensitized and direct photolyses (254 nm) of aerated solutions (pH 9.3) of picloram ($1.1 \times 10^{-3} M$). Photosensitization of picloram was most pronounced with benzoate and benzophenone where 78.7 and 70.6% of the original picloram decomposed, respectively. Since the triplet energy of picloram is 63.5 kcal/mol, as determined from the phosphorescence data, triplet–triplet energy transfer is energetically favored for benzoate and benzophenone. Their triplet energies are 70 and 69 kcal/mol, respectively. Benzene, phenolate, and acetone were less efficient as photosensitizers, probably due to the formation of smaller populations of excited molecules in the triplet states. Turro (1967) reported that excited molecules of benzene underwent very inefficient intersystem crossing (singlet \rightarrow triplet) because of the large energy separation between these two states.

A list of the quantum yields (ϕ) for the disappearance of picloram is shown in Table II. Direct photolysis of picloram (aerated) with 313- and 254-nm light gave rise to yields of

Table I. Percent Photodecomposition of Picloram with 254-nm (λ) Light in Presence of Sensitizer (Aerated)

Sensitizer	Concn, <i>M</i>	Triplet energy (E_T), kcal/mol	% photo-decomposition ^e
Benzene	5.0×10^{-3}	85 ^a	37.6
Phenolate	5.0×10^{-3}	~82 ^c	44.8
Benzophenone	1.0×10^{-3}	69 ^a	70.6
Benzoate	2.2×10^{-3}	70 ^b	78.7
Acetone	5.0×10^{-3}	78 ^a	42.5
Nil (picloram only)	1.1×10^{-3}	63.5 ^d	38.0

^a E_T values of sensitizers came from the monograph by Turro (1967). ^b Data taken from studies by Maria and McGlynn (1970). ^c E_T value is for phenol; E_T for the phenolate anion was not found in the literature. ^d Value determined from phosphorescence measurements of picloram in EPA at 77 K. ^e Values are the means of two or more determinations.

Table II. Quantum Yields (ϕ) for Sensitized Photodecomposition of Picloram with 313- and 254-nm Light^a

Sensitizer	Concn, <i>M</i>	313 nm ^b	254 nm ^b
Benzophenone	$<1.0 \times 10^{-3}$ (aer)	0.110	0.070
	$<1.0 \times 10^{-3}$ (deaer)	0.090	0.060
Benzoate	2.2×10^{-3} (aer)	0.040	0.080
	2.2×10^{-3} (deaer)	0.038	0.078
Nil (picloram only)	1.1×10^{-3} (aer)	0.040	0.037
	1.1×10^{-3} (deaer)	0.039	0.036

^a The measured incident intensities were 8.6×10^{17} quanta sec^{-1} for 313 nm and 3.3×10^{17} quanta sec^{-1} for 254 nm. ^b Quantum yield values are the means of two or more determinations.

0.04 and 0.037, respectively. A quantum yield of 0.005 was reported by Mosier and Guenzi (1973), who photolyzed an aerated picloram solution (2.08×10^{-3} M) (pH 9.5) with light at ~366 nm for 72 hr. The lower quantum yield reported by these investigators probably was due to increased light absorption by secondary products. In the present study, benzophenone increased the quantum yield of the reaction by a factor of nearly 3 with 313-nm light. Triplet-energy transfer from the n, π^* transition of benzophenone at 325 nm (λ_{max}) is believed to be responsible for this sensitization. The lack of sensitization by benzoate with 313-nm light is probably due to the absence of an electronic transition near this frequency. The n, π^* transition of benzoate is not visible, but is hidden under the π, π^* transition, which occurs at 270 nm (λ_{max}).

The role of oxygen in the photochemical reactions of picloram is not completely understood, but the results demonstrate that the rate of photodecomposition is faster in the presence of oxygen. In both direct and sensitized photolyses of the present investigation, the loss of picloram was 4–5% higher in aerated solution. Mosier and Guenzi (1973) reported nearly a 20% increase in the photodecomposition of picloram. No explanation can be made to account for the large discrepancy in the results of the two experiments. These investigators concluded that oxygen was responsible for the photooxidation of radical intermediates.

The present evidence suggests that the triplet state is involved in the primary photochemical process of picloram; however, oxygen and other quenchers do not appear to effectively quench the reaction. Cleavage of strained carbon-chlorine and carbon-carboxyl bonds as a result of steric hindrance created by the triplet-state geometry may ac-

Table III. Percent Yield of $^{14}\text{CO}_2$ Resulting from Sensitized Photodecomposition of Picloram- ^{14}C

Sensitizer	Concn, <i>M</i>	% yield of $^{14}\text{CO}_2$ ^a	
		313 nm	254 nm
Benzophenone	$<1.0 \times 10^{-3}$ (aer)	12.7	15.8
	$<1.0 \times 10^{-3}$ (deaer)	7.4	9.9
Benzoate	2.2×10^{-3} (aer)	8.1	31.5
	2.2×10^{-3} (deaer)	5.0	16.7
Nil (picloram only)	1.1×10^{-3} (aer)	6.0	10.4
	1.1×10^{-3} (deaer)	3.7	6.2

^a Values are means of two or more determinations.

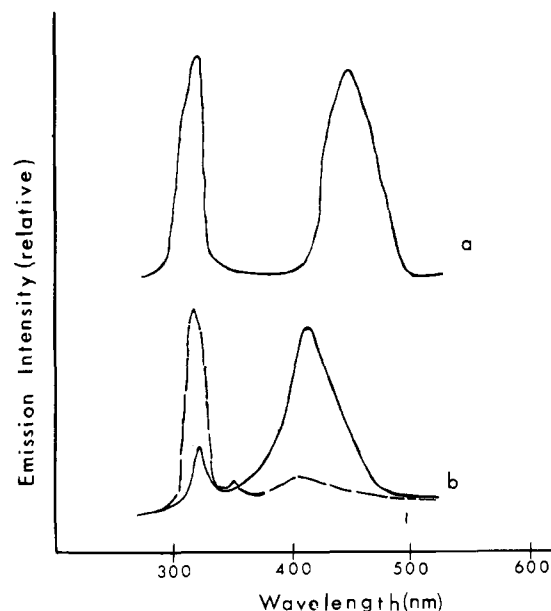


Figure 1. (a) Phosphorescence emission spectrum of picloram (free acid) in EPA glass at 77 K; (b) fluorescence emission spectra (uncorrected) of picloram in aqueous solutions of pH 0.68 (—) and pH 3.44 (---) at room temperature.

count for the initiation of the reaction from the triplet level even in the presence of oxygen.

The percent yields of $^{14}\text{CO}_2$ resulting from the decarboxylation of picloram are shown in Table III. Nearly twice as much $^{14}\text{CO}_2$ was liberated by the 254-nm light than with the 313-nm light and this difference is believed to be caused by the self-absorption of the excitation energy by picloram [λ_{max} 252 nm, $\log \epsilon = 3.17$; λ_{max} 290 nm, $\log \epsilon = 2.67$ (Glass, 1974)]. The photodecarboxylation of picloram was significantly enhanced by the triplet energy sensitizers; benzoate increased the $^{14}\text{CO}_2$ yields (percent) by a factor of about 3 in the photolysis with 254-nm light. These data reveal that the photodecarboxylation of picloram is not the dominant photochemical reaction, but demonstrate that it is a significant pathway.

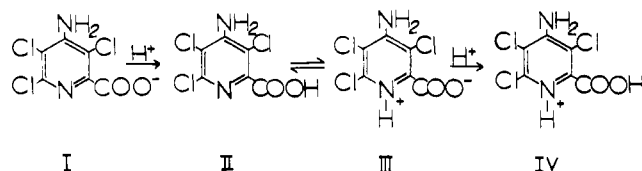
Emission Spectra. The phosphorescence emission spectrum of a solid solution of picloram in EPA at 77 K is shown in Figure 1a. The maximum excitation and emission wavelengths are 320 and 450 nm. Similar results were obtained in ethanol glass at 77 K. The decay times of the phosphorescence for the free acid and the methyl ester of picloram are 40 and 32 msec, respectively. The radiative lifetime indicates that the lowest triplet transition originates from an n, π^* state with an energy equivalent to 63.5 kcal/mol.

The fluorescence excitation and emission wavelengths observed for aqueous solutions of picloram are 320 and 425

Table IV. Effect of pH on the Fluorescence Emission Intensity of Picloram at 425 nm

Sample	pH	Intensity, %	Sample	pH	Intensity, %
1	0.68	100	4	3.44	13
2	1.16	100	5	4.80	10
3	2.00	20	6	9.28	11

nm, as shown in Figure 1b. The data in Table IV show the pH dependence of the fluorescence over the pH range of 0.68 to 9.28. The fluorescence intensity is a maximum from pH 0.68 to 1.16, where picloram is predominantly in its protonated forms (II, III, IV) as reported by Cheung and Biggar (1974). A decrease in fluorescence occurs from 1.68 to 2.0, probably due to quenching by the molecular (II) and/or the dipolar (III) forms. Above pH 4.0, where the anion (I) predominantly exists, the fluorescence is very weak and remains constant through pH 9.28. The pK_a of picloram is 3.4 at ambient temperature.



The fluorescence emission data suggest that the lowest excited triplet of picloram (I, II, III) is an n, π^* state, which usually detracts from the fluorescence of most nitrogen heterocycles (Hercules, 1966). In the protonated species (IV), the n -electrons on the ring nitrogen are removed by protonation. Consequently, this reaction diminishes the influence of the electron pair on the excited state, thus enhancing fluorescence. These results demonstrate that the excitation energy in all molecular forms of picloram, except the protonated, undergoes intersystem crossing to the triplet state at the expense of fluorescence. The electronic configuration remains the same in this process (i.e., $^1n, \pi^* \rightarrow ^3n, \pi^*$).

CONCLUSION

The present results demonstrate that the photosensitization of picloram occurs in the presence of efficient triplet-

energy sensitizers such as benzophenone and benzoate. The phosphorescence emission spectra reveal that photoexcited picloram possesses an n, π^* state equivalent to 63.5 kcal/mol of energy. The quenching of the fluorescence emission by the anions of picloram at pH 9.3 suggests that the excitation energy in the singlet state undergoes intersystem crossing to the triplet state. It is concluded that photodecomposition of picloram takes place from the triplet state along with the emission of phosphorescent light.

ACKNOWLEDGMENT

The author wishes to thank D. L. Martin of the University of Maryland at College Park, M. L. Greenough of the National Bureau of Standards, and R. A. Passwater of AM-INCO, Silver Spring, Md., for allowing the use of their instruments in this investigation, and Dow Chemical Company for supplying the picloram.

LITERATURE CITED

- Bovey, R. W., and Scifres, C. J., "Residual Characteristics of Picloram in Grassland Ecosystems", Texas A&M University, 1971.
- Cheung, M. W., and Biggar, J. W., *J. Agric. Food Chem.* **22**, 202 (1974).
- Glass, B. L., unpublished data, 1974.
- Greenough, M. L., and Hammond, H. K., III, "Technical Manual for Phosphor Standard Calibrator", NBSIR 74-552, National Bureau of Standards, Department of Commerce, Washington, D.C., Oct 1974.
- Hall, R. C., Giam, C. S., and Merkle, M. G., *Weed Res.* **8**, 292 (1968).
- Harbaugh, K. F., O'Donnell, C. M., and Winefordner, J. D., *Anal. Chem.* **45**, 381 (1973).
- Hatchard, C. G., Parker, C. A., *Proc. R. Soc. London, Ser. A* **235**, 518 (1956).
- Hercules, D. M., "Fluorescence and Phosphorescence Analysis", Interscience, New York, N.Y., 1966, p 92.
- Hunt, R. E., and Davis, W., *J. Am. Chem. Soc.* **69**, 1415 (1947).
- Maria, H. J., and McGlynn, S. P., *J. Chem. Phys.* **52**, 3399 (1970).
- Matthews, R. W., and Sangster, D. F., *J. Phys. Chem.* **69**, 1938 (1965).
- Mosier, A. R., and Guenzi, W. D., *J. Agric. Food Chem.* **21**, 835 (1973).
- Turro, N. J., "Molecular Photochemistry", W. A. Benjamin, New York, N.Y., 1967.

Received for review March 26, 1975. Accepted July 23, 1975. Trade names are used for the convenience of the reader and do not constitute any preferential endorsement by the U.S. Department of Agriculture over similar products available.