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Levels of N-Nitrosodimethylamine in Nitrogen Fertilizers/Herbicide Mixtures Containing 2,4-D Present as Dimethylamine Salt

Yuk Y. Wigfield and Charles C. McLenaghan

Laboratory Services Division, Food Production & Inspection Branch, Agriculture
Canada, Ottawa, Ontario, K1A OC6, Canada

The nitrogen content of solid or liquid fertilizers may be present as free ammonia (in liquid fertilizer only), ammonium nitrate or urea. It is suspected that ammonium nitrate may contain nitrite as a trace impurity. In Canada, mixtures of fertilizer and herbicide are commercially available and generally known as "weed and feed" products. Three of the many herbicides used to formulate these products are 2,4-D, mecoprop and dicamba all of which are present as the corresponding dimethylamine (DMA) salts.

DMA and nitrite are the two necessary precursors of N-nitrosodimethylamine (NDMA) which has been found to be carcinogenic to test animals (Seiler 1977; Borzsonyi et al. 1978). Thus a study was initiated to investigate whether NDMA is generated from mixtures of nitrogen fertilizers and herbicides present as the corresponding DMA salts and to determine the levels of NDMA in these mixtures.

Several methods of analysis for NDMA, a volatile nitrosamine, have been reported (Bontoyan et al. 1979; Wigfield et al. 1987; Hindle et al. 1987) including the use of a gas-liquid chromatograph combined with a Thermal Energy Analyser (GC/TEA) or a mass spectrometer (GC/MS). The present paper reports the findings of this investigation using a published cleanup method for N-nitrosodipropylamine in trifluralin formulations (Wotherspoon et al. 1988) and a GC/TEA detection system.

MATERIALS AND METHODS

Standard solutions of NDMA (0.1 mg/mL in ethanol or 0.06 ug/mL in 10% ethanol in dichloromethane) were prepared by diluting the appropriate aliquots of pure NDMA (Sigma, St. Louis, Mo). Absolute alcohol was USP grade, dichloromethane was LC grade and water was purified with a Milli-Q system.

Send reprint requests to Yuk Y. Wigfield at the above address

Silica gel 60 (Merck) was conditioned at 120°C, stored in a desiccator containing drierite and used within 5 days. Sodium sulfate (BDH Chemicals) was Analar grade.

A minicolumn using a disposable Bond Elut tube (3 mL, Analytichem, Harbour City, CA) was prepared as described by Wotherspoon et al. (1988) and was connected to a SPE vacuum manifold with a rack of test tubes (16x125 mm). The column was washed under vacuum with dichloromethane (6 mL).

To analyse NDMA in fertilizers (Table 1), a sample (5 g or 10 g for samples 1 and 5) in a 40 mL diposable centrifuge tube (Becton, Dickinson & Co., Ma) containing water (10 mL) was shaken on a horizontal sample shaker (Eberbach, Ann Arbor, Mich.) and extracted with dichloromethane (2x10 mL). The mixture was centrifuged for 5-10 min or until the two phases were completely separated and the organic phase was added into the minicolumn. The column was eluted with 10% ethanol in dichloromethane (4.5 mL), the last 4 mL of which was collected and diluted to 5 mL with the same solvent system.

Table 1. Fertilizers used to mix with 2,4-D DMA formulation

Sample no.	Available N-P-K(1)(%)	Physical state	Sample no.	Available N-P-K(1)(%)	Physical state
1	17-17-17	granules	10	34-0-0	granules
2	29-0-0	liquid	11	34-0-0	granules
3	NA(2)	granules	12	40-0-0	granules
4	28-0-0	liquid	13	34-0-0	granules
5	14-0-0	granules	14	46-0-0	granules
6	26-0-0	granules			
7	NA	granules			
8	28-0-0	liquid			
9	28-0-0	liquid			

(1) N-P-K = nitrogen-phosphorous-potassium.
(2) NA = not available.

To analyse NDMA in 2,4-D DMA salt (Canadian Pest Control Product No. 11779, 500 g/mL), a sample (1 g) was mixed with water (6 mL) and extracted with dichloromethane (2x3 mL). The extracts were treated in the same manner as those from the fertilizer samples.

To analyse NDMA in a mixture of 2,4-D DMA salt and fertilizer, a sample of fertilizer (5 g or 10 g for samples 1 and 5) was mixed with 2,4-D DMA salt (1 g) and water (10 mL) in a 40 mL disposable centrifuge tube and was stored at room temperature in a dark cupboard (Table 2). After a specific storage time, dichloromethane (10 mL) was added to the mixture and the resulting solution was treated in the same manner as the fertilizer sample.

Aliquots (1 uL) of extracts from the minicolumns were injected into a GC/TEA. The GC column, injection port, helium carrier gas and TEA vacuum were respectively maintained at 90°C, 150°C, 10 mL/min and 1.0 mm Hg. All other instruments and operation parameters were the same as previously described (Wigfield et al.

1987). Under these conditions, the retention time of NDMA was 1.86 min.

RESULTS AND DISCUSSION

In Canada, mixtures of fertilizers and herbicides, generally known as "weed and feed", are popularly used in agriculture and around gardens in urban areas. They can be formulated with various combinations of fertilizers and herbicides. Instead of analysing the actual commercial "weed and feed" products which contain approximately 1% herbicides, it was decided to mix the two ingredients in the laboratory, store the mixture at room temperature for a sufficient duration to allow the formation of NDMA if the reaction conditions were favorable and then analyse the mixture for NDMA. The reasons for so doing are: (a) to be able to obtain a reference level of NDMA already present in the herbicide (0.24 ppm, see Table 2) and (b) to generate a worst-case scenario for the formation of NDMA from the mixture, i.e., using higher weight ratios of fertilizer containing nitrate and herbicide present as DMA salt only. If, under such conditions, generation of NDMA in the mixture is not observed, it can confidently be assumed that the commercial products will not generate NDMA under similar or less favourable conditions.

Accordingly, fourteen solid or liquid fertilizers from the Canadian market, containing available nitrogen in the range of

Table 2. The levels of NDMA in mixtures of fertilizers and 2,4-D DMA formulation

Sample	Weight of mixture (g)		Storage	NDMA
no.(1)	Fertilizer	2,4-D DMA	time(days)	(ppm)(2)
1	10.196	0.505	10	ND(3)
2	5.093	0.526	10	0.19
3	5.000	0.510	10	0.20
4	5.833	0.503	10	0.20
5	10.237	0.503	10	0.22
6	5.107	0.500	10	0.19
7	5.034	0.528	10	0.19
8	5.066	0.512	10	0.27
9	5.755	0.506	7	ND
10	4.993	0.507	7	0.20
11	5.067	0.510	7	ND
12	5.544	5.520	7	0.17
13	5.090	0.517	7	0.22
14	5.755	0.506	7	0.13
	0.0	0.500	7	0.23
	0.0	0.500	0	0.24

(1) Same sample no. as in Table 1.
(2) Based on content of 2,4-D DMA in the mixtures.
(3) ND = Non-detectable with a detection limit of 0.05 ppm.

14-46% (Table 1), were mixed with herbicide containing 500 g/mL 2,4-D DMA salt. The findings of NDMA in the mixtures and in herbicide, expressed in ppm relative to the weight of 2,4-D DMA, are shown in Table 2.

Table 3. Percent recoveries of NDMA from fortified 2,4-D DMA formulation

	NDMA (ppm)			Recovery (%)
	Present(A)	Fortification(B)	Found(C)	(C-A)/B x 100
	0.24	0.59	0.80	95
	0.23	0.61	0.79	92
	0.24	0.59	0.77	90
	0.24	0.59	0.79	93
	0.23	0.57	0.71	84
	0.23	0.59	0.69	78
	0.23	0.32	0.55	100
Average	0.23			90.3
SD (n=7)	0.01			7.3
CV (n=7)	0.02%			8.1

The cleanup method chosen for these samples was modified by Wotherspoon et al. (1988) from a published method (Maybury et al. 1983) for the determination of N-nitrosodipropylamine in trifluralin formulations. The recovery studies conducted in 2,4-D DMA salt (Table 3), fertilizers (Table 4) and mixtures of fertilizers and 2,4-D DMA salt (Table 5), the resulting standard deviation (SD) and coefficient of variation (CV) using standard statistical calculations demonstrate that the method is applicable to these samples.

Table 4. Percent recoveries of NDMA from fortified fertilizers

Sample no.	NDMA (ppm)(1) Present	Fortification(A)	Found(B)	Recovery (%) B/C x 100
1	ND(2)	0.029	0.023	81
		0.026	0.024	89
8	ND	0.060	0.048	80
		0.058	0.051	88
9	ND	0.059	0.048	81
		0.058	0.050	86
11	ND	0.058	0.052	89
		0.059	0.054	91
12	ND	0.060	0.049	81
Average (at 0.06 ppm, n=7)			0.050	85.1
SD (n=7)			0.002	4.45
CV (n=7)			4.43%	5.23

(1) Based on content of fertilizer.

(2) ND = Non-detectable with a detection limit of 0.01 ppm.

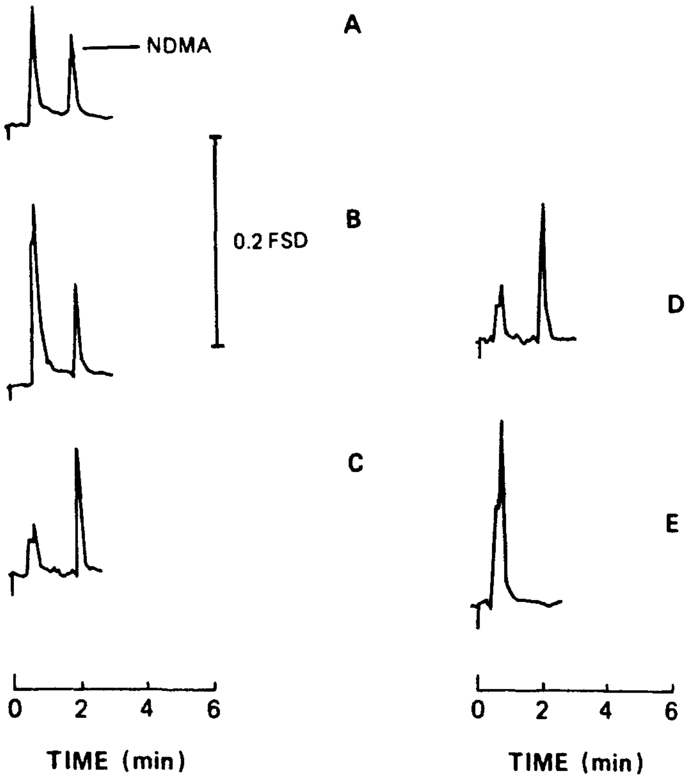

Figure 1. TEA response to NDMA in: (a) herbicide containing 500 g/L 2,4-D DMA salt, (b) mixture of fertilizer sample 8 and herbicide, (c) fertilizer sample 1 spiked with 0.3 ug NDMA, (d) NDMA standard (0.06 ng) and (e) fertilizer sample 1.

Table 5. Percent recoveries of NDMA from fortified mixtures of fertilizers and 2,4-D DMA formulation

Sample no.	NDMA (ppm)(1) Present(A)	Fortification(B)	Found(C)	Recovery (%) (C-A)/B x 100
8	0.24	0.59	0.84	101
	0.24	0.59	0.75	87
	0.20	0.59	0.70	85
	0.20	0.55	0.66	84
12	0.24	0.58	0.74	86
	0.24	0.58	0.72	80
	0.18	0.60	0.72	91
	0.19	0.57	0.71	92
Average				88
SD (n=8)				6.4
CV (n=8)				7.2

(1) Based on content of 2,4-D DMA in the mixtures.

The limit of detection (LOD) for fertilizer samples was defined as 3 x SD of sample spiked at 0.06 ppm and was 0.01 ppm (Table 4). The limit of quantitation (LOQ) was defined as 10 x SD and was 0.02 ppm. Similarly, the values of LOD and LOQ for 2,4-D DMA were 0.02 and 0.05 ppm respectively (Table 3). Figure 1 shows the absence of chromatographic interferences in: (a) 2,4-D DMA formulation (b) mixture of fertilizer (sample 8) and herbicide and (c) fertilizer (sample 1) spiked with NDMA (0.3 ug).

All fertilizer samples were analysed for the presence of NDMA prior to mixing with herbicide using the described method. NDMA was not detected in any of the samples. The identity of NDMA in herbicide and in the mixtures was confirmed by comparison of the TEA response time of a standard solution of NDMA (Figure 1).

Results in Table 2 show that NDMA levels in mixtures of fertilizers and 2,4-D DMA stored at room temperature for 7 to 10 days were within the analytical variation of that originally present in the herbicide. Thus, it can be concluded that mixtures of various fertilizers and 2,4-D DMA salt do not generate any significant additional NDMA.

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