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# Effects of Chemical Modification on the Epoxytrichothecene-Induced Feed Refusal Response

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Six derivatives of the epoxytrichothecene *Fusarium* mycotoxin T-2 toxin were prepared and tested for feed refusal activity by using a mouse drinking-water bioassay. Catalytic hydrogenation of the double bond of T-2 tetraol decreased the biological activity by half. Destruction of the spiro epoxy group by lithium aluminum hydride reduction resulted in elimination of the refusal response. A decontamination procedure consisting of mild alkaline hydrolysis of T-2 toxin to T-2 tetraol followed by a thermal hydration reaction gave a mixture of epimeric hydrates and a tetrahydroxy ketone. These compounds were also devoid of refusal activity.

Feed refusal is one of the characteristic responses produced in swine by the epoxytrichothecene class of mycotoxins. The naturally occurring epoxytrichothecenes T-2 toxin, diacetoxyscirpenol, and vomitoxin (deoxynivalenol) all give rise to the same degree of refusal by swine (Vesonder et al., 1979). The similar response of rodents to these toxins led to development of a simple and rapid bioassay for refusal activity based on the consumption by mice of drinking water containing test compounds (Burmeister et al., 1980). This assay was utilized in the present study to determine which structural features of the epoxytrichothecene T-2 toxin (Figure 1: 1) contribute to or are necessary for feed refusal activity. It has previously been shown that transformations of diacetoxyscirpenol, which open the spiro epoxy ring, result in compounds that lack cytotoxicity (Grove and Mortimer, 1969). Once the structural requirements for feed refusal are known, then suitable processes for detoxification of feedstuffs and other materials containing these mycotoxins can be developed. This paper reports the preparation of six derivatives of T-2 toxin, their evaluation in the mouse drinking-water bioassay, and a procedure for decontamination by thermal hydration.

## EXPERIMENTAL SECTION

**Analytical and General Procedures.** Infrared spectra (IR) were recorded with a Perkin-Elmer 1320 spectrophotometer. Mass spectra (MS) were obtained by chemical ionization with isobutane as the reagent gas, using a Finnigan MAT triple-stage quadrupole mass spectrometer operated in the $Q_3$ mode. Samples were introduced by a direct insertion probe or a gas chromatograph (GC) inlet. For GC–MS, a 3-ft 3% OV-1 column was temperature programmed from 175 to 260 °C at 4 °C/min. The helium flow rate was 20 mL/min and the GC was coupled to the MS by a single-stage glass jet separator. $^{1}H$ and $^{13}C$ nuclear magnetic resonance (NMR) spectra were recorded in $CDCl_3$ at 300 and 75 MHz, respectively, on a Bruker WM-300WB instrument with $Me_4Si$ as the internal standard.

Column chromatographic separations were done on silica gel 60 (70–230 mesh, EM Laboratories). Thin-layer chromatography (TLC) was carried out on 0.25 mm thick layers of silica gel 60, F-254 (EM Laboratories). Components were visualized by spraying with 3% ceric sulfate in 3 N $H_2SO_4$ and heating at 110 °C for 15 min. Acetylations were carried out with acetic anhydride–pyridine (1.5:2.0) at room temperature for 24 h, unless stated otherwise. Reaction mixtures were concentrated to dryness below 40 °C at reduced pressure prior to column chromatography. Melting points were determined with a Mettler FP1 instrument and are uncorrected. Precautions were taken to avoid dermal contact with the epoxytrichothecenes.

**Acetyl T-2 Toxin (2).** T-2 toxin [1 (Burmeister, 1971)] (47 mg, 0.1 mmol) was acetylated, and the product was chromatographed on silica gel (4 g). Elution with dichloromethane–methanol (99:1) gave 50 mg of 2: MS $m/z$ (rel intensity) 509 ($MH^+$, 7), 407 (30), 347 (82), 317 (15), 287 (100).

**T-2 Tetraol (3).** A mixture of 1 (233 mg, 0.5 mmol) and 5% aqueous ammonium hydroxide (15 mL) was sealed and kept at 37 °C for 4 days. Chromatography of the concentrate on silica gel (15 g) eluted with dichloromethane–methanol (9:1) gave 3 (149 mg, 100%): single spot TLC (dichloromethane–methanol, 9:1); MS $m/z$ (rel intensity) 299 ($MH^+$, 7), 281 (75), 263 (17), 251 (18), 233 (50), 215 (100). Acetylation of 3 gave the tetraacetate: GC–MS $m/z$ (rel intensity) 467 ($MH^+$, 10), 407 (43), 377 (7), 365 (6), 347 (100), 317 (20), 305 (9), 287 (85), 275 (17), 257 (36).

**Dihydro T-2 Tetraol (4).** Hydrogenation of 3 (36 mg) with $PtO_2$ (20 mg) in 95% EtOH was carried out at atmospheric pressure for 1 h. The catalyst was removed by filtration, and the filtrate was concentrated. Chromatography of the residue on silica gel (3 g) eluted with dichloromethane–methanol (9:1) gave 4 (28 mg): GC–MS of the trimethylsilyl derivative showed two isomers that exhibited the same fragmentation pattern, $m/z$ 589 ($MH^+$), 499, 409, 385, 367, 319, 287.

**Trichothec-9-ene-3$\alpha$,4$\beta$,8$\alpha$,12$\alpha$,15-pentaol (5).** A mixture of 1 (233 mg, 0.5 mmol) and 1 M lithium aluminum hydride in tetrahydrofuran (6 mL) was heated under reflux for 24 h. Excess reagent and the resultant complex were destroyed by addition of ethyl acetate (1 mL) and 10% aqueous ammonium chloride (0.5 mL). The suspension was filtered through Celite, and the precipitate was washed with methanol. Chromatography of the combined concentrated filtrates on silica gel (10 g) eluted with dichloromethane–methanol (9:1) gave 5 (137 mg, 91%): single spot TLC $R_f$ 0.38 (dichloromethane–methanol, 4:1); MS $m/z$ (rel intensity) 283 ($MH^+ - H_2O$, 45), 265 (75), 253 (100), 247 (60), 235 (100), 229 (15), 223 (88), 217 (92), 205 (65). Acetylation gave the 3,4,8,15-tetraacetate, which was chromatographed on silica gel (4 g) eluted with dichloromethane–methanol (99:1): mp 130–131.5 °C (ether–hexane); IR ($CHCl_3$) 3600, 2970, 1735, 1365 $cm^{-1}$; GC–MS $m/z$ (rel intensity) 409 ($MH^+$ – HOAc, 28), 349 (42), 319 (10), 289 (100), 259 (17), 229 (12), 199 (20); $^{1}H$ NMR $\delta$ 1.01 (s, 3 H, 14-$CH_3$), 1.48 (s, 3 H, 13-$CH_3$), 1.74 (br s, 3 H, 16-$CH_3$), 1.99, 2.03, 2.10, 2.14 (4 s, 12 H, acetate methyls).

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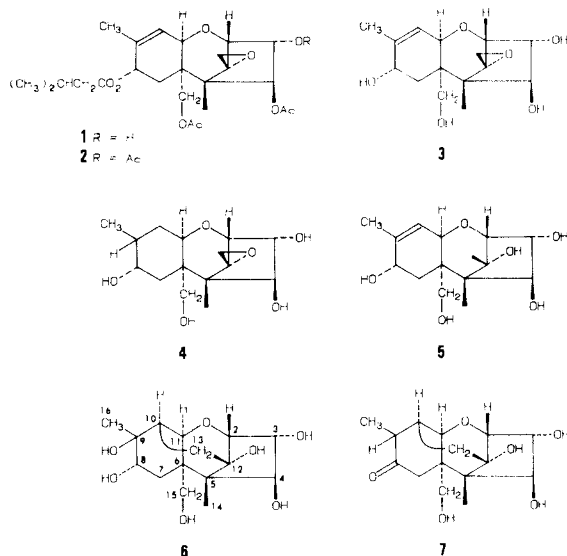


**Figure 1.** Trichothecene derivatives tested for refusal response.

**Hydration of T-2 Tetraol (3).** A solution of **3** (149 mg, 0.5 mmol) in distilled water (12 mL) was heated in an autoclave at 121 °C and 15 psi for 7 h. Chromatography of the concentrate on silica gel (15 g) eluted with dichloromethane–methanol (9:1) gave the ketone **7** (26 mg, 17%): IR (KRS-5 plate) 3380, 1705 $cm^{-1}$; MS $m/z$ (rel intensity) 299 ($MH^+$, 100), 281 (68), 263 (15), 245 (7), 233 (6), 215 (8). Elution with dichloromethane–methanol (85:15) gave the hydrate **6** (111 mg, 70%): MS $m/z$ (rel intensity) 317 ($MH^+$, 50), 299 (100), 281 (30), 263 (15), 245 (7). These compounds exhibited the following $R_f$ values on TLC (dichloromethane–methanol, 4:1): **3**, 0.41; **7**, 0.37; **6**, 0.10 two spots.

**Acetylation of T-2 Tetraol Hydrates (6).** A solution of **6** (74 mg, 0.23 mmol) in acetic anhydride–pyridine was kept at room temperature for 3 days. Chromatography of the concentrate on silica gel (12 g) eluted with dichloromethane–methanol (98:2) gave the 3,4,8,15-tetraacetate A (53 mg, 48%): mp 274.5–279 °C (acetone–hexane); IR ($CHCl_3$) 3590, 2980, 1742, 1375 $cm^{-1}$; GC–MS $m/z$ (rel intensity) 485 ($MH^+$, 100), 467 (7), 425 (36), 407 (31), 383 (42), 365 (90), 347 (27), 323 (31), 305 (69); $^1H$ NMR $\delta$ 1.13 (s, 3 H, 14-$CH_3$), 1.18 (s, 3 H, 16-$CH_3$), 2.10, 2.11, 2.13 (12 H, acetate methyls), 3.77 (d, 1 H, H-11, $J$ = 3.7 Hz), 3.90 (d, 1 H, H-$15_B$, $J$ = 11.8 Hz), 4.13 (d, 1 H, H-2, $J$ = 4.2 Hz), 4.54 (d, 1 H, H-$15_A$, $J$ = 11.8 Hz), 5.0 (m, 2 H, H-3 and H-8), 5.40 (d, 1 H, H-4, $J$ = 2 Hz); $^{13}C$ NMR $\delta$ 10.3, 20.9, 21.0, 23.9, 27.6, 32.8, 43.5, 46.5, 51.3, 68.2, 71.0, 72.3, 73.5, 78.4, 78.8, 80.8, 82.3, 169.1, 170.3, 170.4, 171.1. Elution with dichloromethane–methanol (95:5) gave the 3,4,8,15-tetraacetate B (36 mg, 32%): mp 210.5–212 °C (acetone–hexane; IR ($CHCl_3$) 3600, 2950, 1742, 1375 $cm^{-1}$; GC–MS $m/z$ (rel intensity) 485 ($MH^+$, 100), 467 (61), 425 (47), 407 (42), 383 (43), 365 (86), 347 (45), 323 (26), 305 (100); $^1H$ NMR $\delta$ 1.16 (s, 3 H, 14-$CH_3$), 1.20 (dd, 1 H, H-$7_B$, $J$ = 14.8, 12 Hz), 1.25 (s, 3 H, 16-$CH_3$), 1.90 (dd, 1 H, H-$13_B$, $J$ = 14.7, 11.9 Hz), 2.06 (dd, 1 H, H-$13_A$, $J$ = 14.7, 5.2 Hz), 2.11, 2.12, 2.13 (12 H, acetate methyls), 2.30 (dd, 1 H, H-$7_A$, $J$ = 14.8, 5.5 Hz), 2.42 (m, 1 H, H-10), 3.38 (d, 1 H, H-11, $J$ = 3.7 Hz), 3.92 (d, 1 H, H-$15_B$, $J$ = 11.6 Hz), 4.17 (d, 1 H, H-2, $J$ = 4 Hz), 4.51 (d, 1 H, H-$15_A$, $J$ = 11.6 Hz), 5.01 (dd, 1 H, H-3, $J$ = 4, 2 Hz), 5.14 (dd, 1 H, H-8, $J$ = 12, 5.5 Hz), 5.41 (d, 1 H, H-4, $J$ = 2 Hz); $^{13}C$ NMR $\delta$ 9.9, 20.8, 20.9, 21.1, 21.6, 26.9, 33.9, 45.4, 46.6, 51.5, 70.0, 70.4, 73.2, 74.7, 78.0, 78.9, 80.9, 81.7, 169.6, 170.5, 171.0, 171.6.

**Acetylation of Ketone 7.** A solution of **7** (32 mg, 0.11 mmol) in acetic anhydride–pyridine was kept at room temperature for 24 h. Chromatography of the concentrate on silica gel (5 g) eluted with dichloromethane–acetone (9:1) gave the triacetate **8** (29 mg, 62%): IR ($CHCl_3$) 3590, 1740, 1715 sh, 1370 $cm^{-1}$; GC–MS $m/z$ (rel intensity) 425 ($MH^+$, 70), 365 (100), 347 (25), 323 (18), 305 (40), 263 (40); GC–MS of the trimethylsilyl derivative $m/z$ (rel intensity) 497 ($MH^+$, 28), 437 (80), 377 (100); $^{13}C$ NMR $\delta$ 10.2, 16.6, 20.7, 20.8, 31.2, 39.8, 42.8, 49.6, 51.5, 52.3, 67.1, 69.4, 77.9, 78.4, 81.0, 81.1, 169.3, 170.3, 170.6, 212.2.

**Acetylation of Ketone Triacetate 8.** A solution of **8** (13.7 mg, 0.03 mmol) in acetic anhydride–pyridine was kept at 45 °C for 2 days. Chromatography of the concentrate on silica gel (2 g) eluted with dichloromethane–acetone (95:5) gave the enol tetraacetate **9** (10.4 mg, 69%): single spot TLC $R_f$ 0.24 (chloroform–acetone, 85:15); IR ($CHCl_3$) 3600, 1740, 1365 $cm^{-1}$; GC–MS $m/z$ (rel intensity) 467 ($MH^+$, 13), 407 (8), 365 (10), 347 (8), 305 (100), 287 (4), 263 (5); GC–MS of the trimethylsilyl derivative $m/z$ (rel intensity) 539 ($MH^+$, 25), 479 (18), 437 (19), 419 (7), 377 (100); $^1H$ NMR $\delta$ 1.01 (s, 3 H, 14-$CH_3$), 1.07 (d, 3 H, 16-$CH_3$, $J$ = 7 Hz), 2.07, 2.08, 2.11, 2.12 (12 H, acetate methyls), 3.54 (d, 1 H, H-11, $J$ = 2.8 Hz), 3.96 (d, 1 H, H-$15_B$, $J$ = 11.2 Hz), 4.15 (d, 1 H, H-2, $J$ = 3.6 Hz), 4.56 (d, 1 H, H-$15_A$, $J$ = 11.2 Hz), 5.09 (dd, 1 H, H-3, $J$ = 3.6, 2.8 Hz), 5.22 (s, 1 H, H-7), 5.41 (d, 1 H, H-4, $J$ = 2.8 Hz); $^{13}C$ NMR $\delta$ 10.2, 17.4, 20.8, 20.9, 21.0, 31.4, 38.7, 39.6, 47.3, 51.7, 66.6, 67.0, 77.7, 80.1, 81.3, 117.3, 151.0, 169.5, 169.7, 170.6, 171.0.

**Sodium Borohydride Reduction of Ketone 7.** A solution of **7** (27 mg, 0.09 mmol) and sodium borohydride (20 mg) in distilled water was kept at room temperature for 24 h. The reaction mixture was acidified with 2 N $H_2SO_4$ and concentrated to dryness. Chromatography of the concentrate on silica gel (3 g ) eluted with dichloromethane–methanol (9:1 and 85:15) separated two isomeric pentaols **10A** (9 mg, 33%) and **10B** (12 mg, 44%): MS $m/z$ (rel intensity), for A, 301 ($MH^+$, 17), 283 (100), 265 (22), 247 (14), and, for B, 301 (52), 283 (100), 265 (39), 247 (16).

**Mouse Drinking-Water Bioassay.** A modification of the previously described bioassay was employed (Burmeister et al., 1980). Fourteen young adult Hsd (ICR) Swiss female mice 7–10 weeks of age were used for each test. Compounds were dissolved in 2% ethanol in distilled water at a concentration of 50 mg/L. An equal number of control mice received only 2% ethanol in water. Volumes of drinking solutions consumed after 20 h were measured. The volume of test solution consumed expressed as a percentage of control consumption subtracted from 100 equals the refusal response.

## RESULTS AND DISCUSSION

T-2 toxin (**1**) was selected as the starting mycotoxin for this study for three reasons: quantities are readily produced by laboratory fermentation (Burmeister, 1971); it is one of only four epoxytrichothecenes known to occur naturally (Pathre and Mirocha, 1977); it is one of the more lethal mycotoxins of this class (Sato and Ueno, 1977). Initially, four derivatives of T-2 toxin were prepared by standard chemical reactions. The 3-hydroxyl of **1** was acetylated to give acetyl T-2 toxin (**2**), which is reported to have much less emetic activity than **1** in a pigeon assay (Kotsonis et al., 1975). Hydrolysis of the three ester groups of **1** was carried out with 5% aqueous ammonium hydroxide to give a quantitative conversion to T-2 tetraol (**3**), which was characterized by chemical ionization mass spectrometry (CI-MS) and by conversion to the tetraacetate. Catalytic hydrogenation of **3** gave 9,10-dihydro T-2 tetraol (**4**), which consisted of a mixture of two hydrogenation isomers as shown by GC–MS of the tri-

methylsilyl derivative. Treatment of 1 with lithium aluminum hydride resulted in reductive cleavage of the three ester groups and the spiro epoxide to give the pentaol **5**. CI-MS of **5** and its 3,4,8,15-tetraacetate failed to show $MH^+$ ions. Instead, the highest mass ions observed represented facile eliminations of water and acetic acid, respectively, from the parent molecules. However, the presence of four acetate methyls was apparent from the $^1H$ NMR spectrum of acetylated **5**. In addition, the AB quartet characteristic for the C-13 epoxy methylene was replaced by a new methyl singlet at $\delta$ 1.48.

It has been reported that treatment of the epoxytrichothecenes diacetylverrucarol and triacetoxyscirpenol, which lack substitution at C-8, with boiling water results in epoxide ring opening involving participation of the 9,10 double bond to give hydrates in about 50% yield (Gutzwiller et al., 1964; Sigg et al., 1965). Treatment of T-2 toxin (**1**) under these conditions resulted in recovery of the starting material. Similarly, treatment of **1** with boiling glacial acetic acid failed to produce an acetic acid adduct but instead gave acetyl T-2 toxin (**2**) plus recovered **1**. However, T-2 tetraol (**3**) did partially react with boiling water to give more polar compounds. Subsequently, treatment of **3** with water in an autoclave at 121 °C and 15 psi for 7 h resulted in nearly complete consumption of **3**. Partial separation of the reaction mixture by column chromatography afforded the major and lowest $R_f$ product, which consisted of two compounds as shown by TLC. CI-MS of this mixture showed a $MH^+$ ion at $m/z$ 317, which indicated formation of hydrate **6** in 70% yield. In order to more completely characterize the hydrate, acetylation of **6** gave a mixture of isomeric acetates A and B in the ratio of 1.5:1, which were separable by column chromatography. Both acetates exhibited a free hydroxyl in their IR spectra. Upon GC–MS analysis, they produced nearly identical fragmentation patterns characterized by losses of water and acetic acid, the only notable difference being that the $MH^+ - H_2O$ ion from the more volatile (GC) and less polar (TLC) tetraacetate A was only 7% of the intense $MH^+$ ion, whereas this ion from B was 61% of the $MH^+$ ion. A two-dimensional chemical shift correlation spectroscopy $^1H$ NMR experiment performed on B allowed assignments to be made for all the ring protons (see Experimental Section). $^1H$ NMR spectra of the two 3,4,8,15-tetraacetates are very similar, with the major difference being the chemical shift of H-11 at $\delta$ 3.77 in A and $\delta$ 3.38 in B. Instead of a vinyl methyl signal at $\delta$ 1.7, the 16-methyls appeared as singlets at $\delta$ 1.18 and $\delta$ 1.25, respectively. The AB quartet for the spiro epoxy methylene and an H-10 vinyl hydrogen were absent. The $^{13}C$ NMR spectra showed four acetate carbonyl carbons at $\delta$ 169–172. A signal at $\delta$ 33 in the hydrate acetates that is not present in spectra of typical epoxytrichothecenes (Ellison and Kotsonis, 1976) was assigned to the C-13 methylene. These data indicate that the acetates from hydrate **6** are epimers that differ only in their configuration at C-9.

The next most abundant product from the hydration reaction, which was obtained in 17% yield and migrated on TLC just below starting **3**, was characterized as the tetrahydroxy ketone **7**. CI-MS of **7** gave an intense $MH^+$ ion at $m/z$ 299, which indicated the compound is isomeric with **3**; however, the presence of a carbonyl group was apparent from the IR spectrum. The major product of room temperature acetylation of **7** was the ketone triacetate **8** (Figure 2), whose CI-MS showed a $MH^+$ ion at $m/z$ 425; the $^{13}C$ NMR spectrum contained three ester carbonyl carbons at $\delta$ 169–171 and one ketone carbonyl

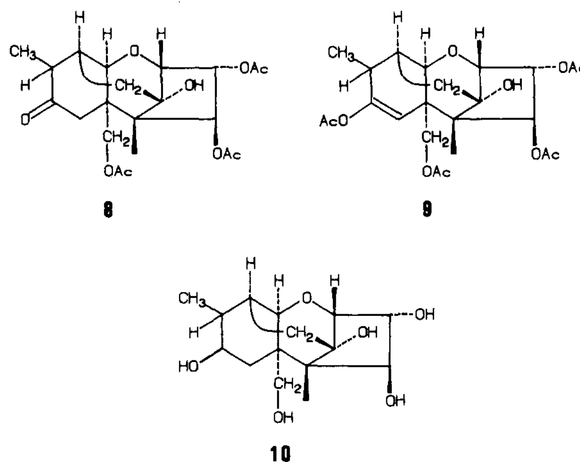


**Figure 2.** Derivatives of tetrahydroxy ketone **7**.

Table I. Refusal Response of Mice to Trichothecene Derivatives

| compound[a] | % drinking solution refused |
|---|---|
| control[b] | 0 |
| T-2 toxin, **1** | 71 |
| acetyl T-2, **2** | 52 |
| T-2 tetraol, **3** | 53 |
| dihydro T-2 tetraol, **4** | 23 |
| LAH product, **5** | 0 |
| hydrates, **6** | 0 |
| ketone, **7** | 1 |

[a] Compound offered in 2% $EtOH-H_2O$ at 50 mg/L to 14 mice. [b] Controls consisting of 14 mice for each test received 2% $EtOH-H_2O$.

carbon at $\delta$ 212.2. In addition, the $^{13}C$ NMR spectrum showed the C-16 methyl at $\delta$ 16.6 and the C-13 methylene at $\delta$ 31.2. The only exceptional features of the $^1H$ NMR spectrum of **8** were the presence of the C-16 methyl as a doublet at $\delta$ 1.2 and the absence of H-8. Trimethylsilylation of **8** produced a mono-$Me_3Si$ derivative ($MH^+$, 497) that confirmed the presence of one tertiary hydroxyl. Further acetylation of **8** at elevated temperature resulted in formation of the enol tetraacetate **9** ($MH^+$, $m/z$ 467), which still possessed one free hydroxyl ($Me_4Si$, $MH^+$, $m/z$ 539). The $^1H$ NMR spectrum showed the C-16 methyl as a doublet at $\delta$ 1.07, four acetate methyls, and a new one-proton singlet at $\delta$ 5.22 assigned to H-7. The $^{13}C$ NMR spectrum confirmed the presence of four ester carbonyl carbons but lacked the ketone carbon. Instead, two new olefinic carbons present at $\delta$ 117.3 and 151.0 were assigned to C-7 and C-8 of **9**, respectively. Final evidence in support of structure **7** was obtained by sodium borohydride reduction of **7** to a mixture of isomeric pentaols **10** ($MH^+$, $m/z$ 301).

Ketone **7** does not arise from the hydrate **6**, which is stable under the hydration conditions. At these elevated temperatures, T-2 tetraol presumably assumes a conformation whereby the 9,10 $\pi$-electrons can attack the epoxide ring (Doyle and Bradner, 1980). The intermediate carbonium ion then either accepts hydroxyl from the solvent to give the major product **6** or is transformed to **7** via an enol.

The results of testing compounds **1**–**7** for refusal activity in the mouse drinking-water bioassay are given in Table I. Compounds were offered to mice in their drinking water at a concentration of 50 mg/L (ppm) over a 20-h period. T-2 toxin (**1**) possesses potent refusal activity and served as the positive control. Either acetylation of the free hydroxyl of **1** or hydrolysis of the three ester groups gave compounds **2** and **3**, respectively, which exhibited slightly

diminished activity. The dihydro derivative 4, which still has the spiro epoxy group, showed about half the activity of the parent tetraol 3. The most striking results, however, were obtained with compounds 5–7 in which the epoxide had been opened. These compounds were consumed at the same rate as the control solution. Even at 100 mg/L, the mixture of epimeric hydrates 6 failed to elicit a refusal response. These results generally parallel those obtained in cytotoxicity tests of similar derivatives from diacetoxyscirpenol (Grove and Mortimer, 1969). In addition, compound 5 has been shown to lack both topical and intraperitoneal toxicity to rats (Bamburg, 1972). Therefore, in terms of feed refusal and perhaps other toxicological properties as well, thermal hydration of 8-hydroxyepoxytrichothecenes offers an approach to decontamination of materials containing these toxins. T-2 toxin, which has a bulky ester substituent at position 8, requires a preliminary mild hydrolysis step to remove this group prior to hydration. The hydration approach may have potential applicability toward a detoxification process, because no hazardous or difficult to remove reagents need to be added to destroy the spiro epoxide ring.

**Registry No.** 1, 21259-20-1; 2, 21259-21-2; 3, 34114-99-3; 4, 89121-46-0; 5, 89121-47-1; 6, 89121-48-2; 6 tetraacetate, 89121-53-9; 7, 89121-49-3; 8, 89121-50-6; 9, 89121-51-7; 10, 89121-52-8.

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# Mass Spectrometry of Cytochalasin H and Its Acetyl and Deacetyl Analogues

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A study was made of the electron ionization (EI) and chemical ionization (CI) (reagent gases: methane and ammonia) mass spectra of cytochalasin H and some of its analogues. The purpose of this work was to characterize these compounds through a study of their mass spectra under EI and CI conditions. The mass spectra are shown to contain all of the information necessary to determine the degree of acetylation as well as the presence of an acyl group on the nitrogen atom. Losses can be followed, accounting for all hydroxyl or acetyl groups on carbons 7, 18, and/or 21 and for an acyl group on the nitrogen atom.

The cytochalasins, metabolites generated by a variety of molds found growing on cereal crops, produce unusual biological effects including inhibition of cell movement, inhibition of cytoplasmic cleavage, and nuclear extrusion of cultured cells. Some cytochalasins have been reported to be acutely toxic to experimental animals (Natori, 1977). In addition, several have been shown to exhibit plant growth inhibitory activity in wheat coleoptiles (Cole et al., 1981; Cutler et al., 1980). During the course of investigation of the plant growth regulating effects of acetate derivatives of cytochalasin H (Beno et al., 1977; Cole et al., 1981; Patwardhan et al., 1974; Wells et al., 1976), it became apparent that the electron impact mass spectra of some of the acetate derivatives did not yield abundant molecular ions with large enough intensity to be useful for characterization purposes (Cox et al., 1983). In order to characterize these cytochalasins more fully, the mass spectral properties of cytochalasin H and its acetyl and deacetyl analogues (Figure 1, 1–5) were investigated further. We report here the results of our study of their electron ionization (EI) and chemical ionization (CI) mass spectra. The mass spectral examinations of the cytochalasin H series of compounds were facilitated by using the technique of desorption chemical ionization (DCI).

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EXPERIMENTAL SECTION

The isolation, purification, and structure elucidation of the cytochalasin H series of compounds has been described previously by Cox et al. (1983).

Chemical ionization and electron ionization mass spectra were obtained by using a Varian MAT 112S mass spectrometer coupled with a Varian MAT SS-200 data system. [The mass spectra under EI and CI conditions are available

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