Synthesis and Biological Evaluation of a Trichothecene *epi*-Epoxide, 3α,4β,15-Triacetoxy-12,13-*epi*-epoxytrichothec-9-ene

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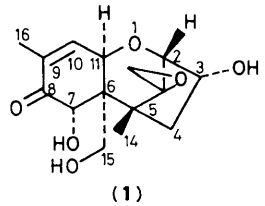
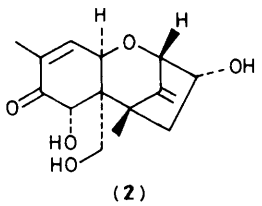
In order to provide additional insight into the mode of action of the trichothecene mycotoxins, one of the first semi-synthetic trichothecene *epi*-epoxides (**8**) has been prepared; in dramatic contrast to its natural isomer (**9**), this compound proved to be devoid of significant biological activity.

The trichothecenes[1] are a group of some 70 complex fungal sesquiterpenoids, some of which are phytotoxic and all of which show some degree of mammalian toxicity. Their fungal occurrence is ubiquitous, and they have been strongly implicated in natural intoxications of both humans and animals. For example, deoxynivalenol (vomitoxin) (**1**) is produced[2] when cereal grains are infected with *Fusarium* species: consumption of feedstuffs contaminated with this toxin can cause sub-lethal toxicoses in animals.

The determination of the tolerance level and the biological pathway of these toxins is of major importance, yet their exact metabolic fate when ingested is as yet incompletely understood.[3,4] Evidence exists which suggests that their toxicity in general does require the presence of the 12,13-epoxide function; these studies[5] showed that biological activity was lost when this function was destroyed, but only by demonstrably deep-seated alterations to the molecule. More recently, experiments with rats *in vivo*[2] and with rumen micro-organisms *in vitro*[6] have shown that the predominant biological transformation of deoxynivalenol (**1**) is one of deoxygenation of the epoxide moiety to form the 9,12-diene (**2**).



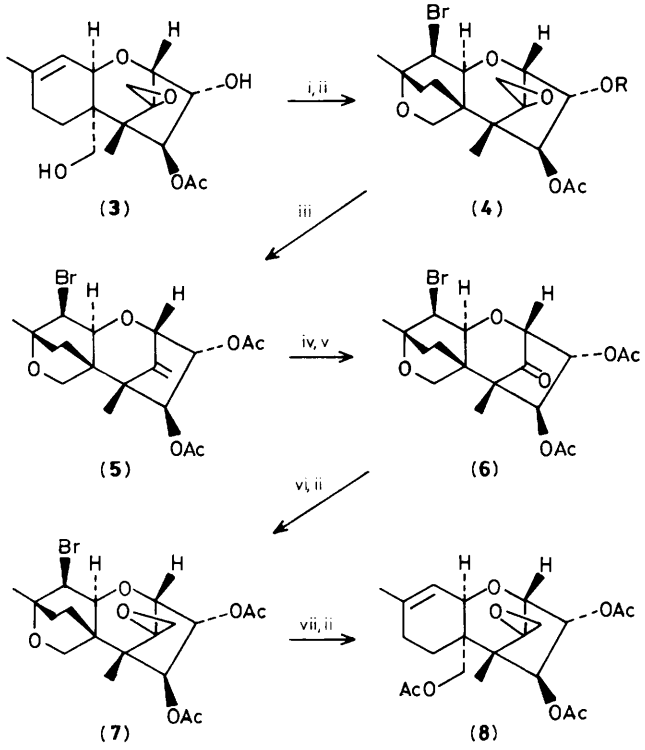
(**1**)



(**2**)

We now communicate the synthesis, from a natural trichothecene, of 3α,4β,15-triacetoxy-12,13-*epi*-epoxytrichothec-9-ene† (**8**); this is one of the first trichothecene

† All reported compounds were fully characterised by elemental analysis and/or high resolution mass spectrometry, and i.r., and ^1H and ^{13}C n.m.r. spectroscopy.

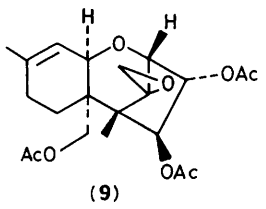


Scheme 1. *Reagents:* i, *N*-bromosuccinimide, MeCN; ii, Ac_2O, pyridine; iii, WCl_6 (3 equiv.), Bu^nLi (7.5 equiv.), tetrahydrofuran (THF), reflux, 4 h; iv, O_3, dichloromethane; v, Et_3N; vi, dimethylsulphonium methylide, THF, 1 h; vii, Zn(Ag), THF, EtOH, Et_2O, reflux, 12 h.

analogues to be prepared with the 'unnatural' epoxide configuration.[7] When compared with its natural isomer (**9**), it proved to be devoid of significant toxicity.

The synthetic sequence is outlined in Scheme 1. 4β-Acetoxy-3α,15-dihydroxy-12,13-epoxytrichothec-9-ene (**3**) was converted[8,9] into the bromo-ether (**4**, R = H), thus affording protection to the 9,10-double bond. Treatment of the derived diacetate (**4**, R = Ac) with the Sharpless[10] tungsten-based deoxygenating system, following a protocol we have successfully applied[11] to deoxynivalenol and anguidine acetates, provided the 12-ene (**5**), in 98% yield. Ozonolysis followed by reductive work-up with triethylamine gave the nor-ketone (**6**). Treatment of this ketone with excess of dimethylsulphonium methylide,[12] followed by reacetylation, gave the epoxide (**7**). Reductive regeneration of the 9,10-double bond using a zinc–silver couple[8] followed by acetylation gave the triacetate (**8**). The 'unnatural' epoxide configuration attained by this route was expected by analogy with earlier observations made in the total synthesis of trichodermin,[13] and was confirmed, *inter alia*, by the appearance in the ^1H n.m.r. spectrum of (**8**) of the AB spin system for the C-13 methylene protons at δ 2.5 and 2.83, *J* 5 Hz: the isomeric, natural trichothecene (**9**) shows the equivalent resonances at δ 2.75 and 3.05, *J* 4 Hz.

Biological evaluation[14] was carried out using human epithelial cells by determining the minimum inhibitory concentration for cell growth. This showed that the *epi*-epoxide (**8**)



was essentially non-toxic, possessing less than one eight hundredth of the activity of its natural isomer (**9**). This finding emphasises the key role played by a correctly orientated epoxide group in providing the cytotoxicity of the trichothecenes.

We thank the S.E.R.C. and the Ministry of Agriculture, Fisheries and Foods (MAFF) for the award of a CASE studentship, Mrs. P. Tait (Glasgow) for mycological assistance, Professor W. R. Roush for providing results prior to publication, Professor B. W. Bycroft (Nottingham University) for advice and a culture for (**3**), and Drs. J. Gilbert and M. Shepherd (MAFF, Norwich) for biological evaluation.

Received, 4th August 1986; Com. 1108

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7 While this manuscript was in preparation, Professor W. R. Roush (Massachusetts Institute of Technology) informed us of his independent degradation studies which have led to syntheses of the trichothecenes analogues 12-*epi*- and 12,13-deoxy-12,13-methanoanguidine, and also of [13-^{14}C]anguidine (W. R. Roush and S. Russo-Rodriguez, *J. Org. Chem.*, in the press).
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