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CHEMISTRY OF INSECT ANTIFEEDANTS FROM AZADIRACHTA INDICA (Part 3)[1]: REACTIONS ON THE C-22,23 ENOL ETHER DOUBLE BOND OF AZADIRACHTIN AND CONVERSION TO 22,23-DIHYDRO-23-β-METHOXYAZADIRACHTIN.

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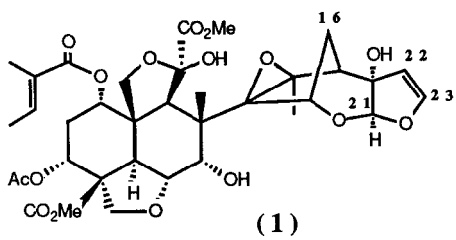
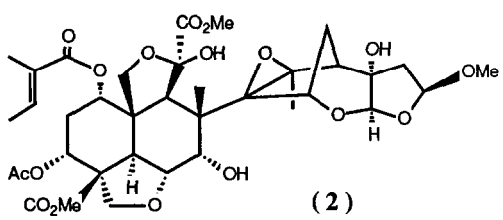
Summary: Azadirachtin (1) can be converted to the natural product 22,23-dihydro-23β-methoxyazadirachtin (2) *via* selective bromomethoxylation of the C-22,23 enol ether double bond and tri-n-butyltin hydride reduction; the corresponding acetic acid adduct on pyrolysis affords (1) in high yield. The antifeedant effects of the addition compounds were assessed.

Azadirachtin (1)[2] is a highly functionalised tetranortriterpenoid isolated from the neem tree *Azadirachta Indica* A. Juss and having potent biological activity as an insect antifeedant and ecdysis inhibitor[3]. For some time we have been following a systematic and integrated research programme to define the structure-activity relationships of azadirachtin[4] and related derivatives[1,5]. Additionally these studies assist in the design of a total synthesis of azadirachtin and related molecules.

In this letter we describe some high yielding modifications to the highly reactive C-22,23 enol ether double bond. The only reaction reported to date of the C-22,23 double bond involves hydrogenation,[2b] which may be accomplished selectively without further reduction of the tigloyl group.



(1)

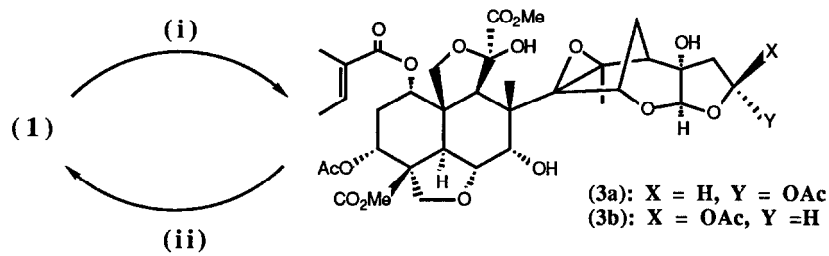


(2)

Although rearrangement of 22,23-dihydroazadirachtin is a facile process[4], the corresponding rearrangement of (1) itself is complicated by side reactions of the enol ether. During investigations of this process it was discovered that (1) underwent a smooth reaction when stirred in acetic acid at room temperature for 3 days. The (2:1) mixture of products (3a,b) (95%), (Scheme 1), could be readily separated by flash chromatography on silica gel.[1]H n.m.r. spectroscopy clearly showed that both of the products resulted from addition of acetic acid across the C-22,23 bond uncomplicated by any side-reaction (Table 1). On the basis of coupling constant measurements the major product (less polar) was assigned as the α-acetoxy compound (3a) and the minor product as β-acetoxy species (3b). More importantly from the synthetic point of view, and quite remarkably, we found that pyrolysis of neat 23-α,β-acetoxy-22,23-dihydroazadirachtin (3), under high vacuum, afforded azadirachtin (1) in essentially quantitative yield.

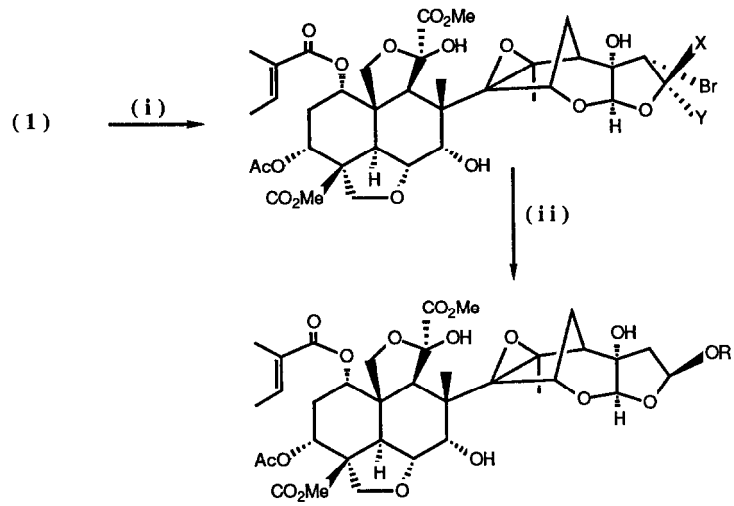
Table 1. Selected 500 MHz ^1H n.m.r. data ($CDCl_3$, $CHCl_3$ = 7.26 p.p.m.) of compounds (1), (3) and (4).

	(1)	(3a)	(3b)	(4a) major isomer
16b	1.31 (d, 13.0)	2.02 (d, 14.0)	2.02 (d, 14.1)	2.01 (d, 10.8)
21	5.65 (s)	5.55 (s)	5.64 (s)	5.76 (s)
22	5.05 (d, 2.0)	2.28 (d, 14.9)	2.36 (dd, 2.8, 15.3)	4.38 (d, 5.8)
	-	2.45 (dd, 5.6, 14.9)	2.54 (dd, 6.8, 15.3)	-
23	6.46 (d, 2.9)	6.38 (d, 5.5)	6.45 (dd, 2.7, 6.8)	5.14 (d, 5.8)
23-OAc	-	2.08 (s)	2.10 (s)	-
23-OMe	-	-	-	3.53 (s)



Scheme 1 (i) AcOH, RT, 72h. (ii) $1.7x10^{-3}$mmHg, 175°C, 5 mins.

Dropwise addition of a solution of bromine in methanol to a solution of (1) in methanol cooled to 0°C, followed by quenching (sodium metabisulphite) and work up, gave a (3:1) mixture of adducts (4a,b) (96%), (Scheme 2). The fast atom bombardment mass spectrum (thiodiethanol) showed molecular ions m/z 853 (MNa^+) and 831 (MH^+), while the high resolution electron impact spectrum showed a strong peak at m/z 361.0274 ($C_{14}H_{18}O_6Br$ requires 361.0287), indicating that addition had occurred to the C-22,23 double bond[2b]. The relative stereochemistry of these two products was established by ^1H n.m.r. spectroscopy and selected one dimensional nuclear Overhauser effect (n.O.e) difference spectra. For the major isomer (more polar) the existence of a H-21/H-23 and a strong H-22/H-16 n.O.e. unequivocally defines the 23-methoxy β and the 22-bromine α (4a). In the case of the minor isomer (less polar) a H-21/MeO-23 n.O.e. replaces the above H-21/H-23 enhancement and the existence of a H-22/H-23 n.O.e. determines the 23-methoxy and 22-bromine both α (4b). Pure 22-α-bromo-22,23-dihydro-23-β-methoxyazadirachtin (4a), obtained by careful flash chromatography, was treated with tri-n-butyltin hydride in boiling benzene containing a trace of azoisobutyronitrile (AIBN) to give the natural product 22,23-dihydro-23,β-methoxyazadirachtin (2) in 82% yield after chromatography. The ^1H n.m.r. spectrum and optical rotation were identical in all respects to the literature data[6] for (2).



(4a): X = OMe, Y = H
(4b): X = H, Y = OMe
(5a): X = OEt, Y = H
(5b): X = H, Y = OEt
(6a): X = O^{i}Pr, Y = H
(6b): X = H, Y = O^{i}Pr

(2) R = Me
(7) R = Et
(8) R = ^{i}Pr

Scheme 2: (i) Br_2, ROH, 0°C, 5 min. (ii) $^{n}Bu_3SnH$, AIBN, Benzene, 80°C.

The corresponding bromo ethoxy (5a,b) (97%) and bromo isopropoxy (6a,b) (96%) derivatives were also prepared along with their corresponding tri-n-butyltin hydride reduced products, the 23-β-ethoxy compound (7) (69%) and the 23-β-isopropoxy compound (8) (68%).

The biological screening for insect antifeeding activity of these compounds were evaluated. Bioassays were undertaken on final stadium larvae of two major pest species of Lepidoptera; *Spodoptera littoralis* (Boisduval) and *Heliothis virescens* (F.). The insects were reared on a bean-based diet[7] at Birkbeck College, at 26±1°C in a 16L:8D photoperiod. The compounds were assessed for antifeedant activity by presenting them on glass fibre discs (Whatman GF/A 2.1 cm diameter) to individual larvae. The discs were made palatable by the addition of 100µl of sucrose (0.05M). The test compounds were dissolved in 90% ethanol and tested at two concentrations, 1ppm and 10ppm. Larvae were placed individually in a Petri-dish with two discs, one of which, the control disc (C), had only sucrose added, the other, the treatment (T) disc, had sucrose and a 100µl aliquot of one of the test solutions. The discs were dried, weighed then presented in pairs (C vs T) to the larva. The duration of the bioassay varied between species but was never longer than 24h, so that never more than 50% of any disc was eaten. The discs were then reweighed and the Antifeedant Index ((C-T)/(C+T)%) calculated on the amounts eaten. This Index identifies both phagostimulants (-ve values) and antifeedants (+ ve values), values in excess of 50 have significant antifeedant activity. Compounds (1), (2) and (7) are potent antifeedant against both species, whereas compounds containing greater steric bulk at C-23 have reduced activity and those containing a further bromine substituent at C-22 are poor antifeedants.

Bioassay Results Antifeedants Index ((C-T)/(C+T)%) [mean* (±SEM)]

Compound	S. littoralis		H. virescens	
Conc./ppm	10	1	10	1
(1) Aza-D	100 ± 0.0	99 ± 1.1	99 ± 1.6	77 ± 4.8
(2) 23-β-OMe	98 ± 1.4	81 ± 3.4	79 ± 4.7	57 ± 6.8
(7) 23-β-OEt	52 ±13.0	66 ± 8.3	59 ±15.0	64 ± 9.8
(3) 23-α,β-OAc	47 ±15.0	49 ± 9.6	38 ± 6.0	39 ± 8.5
(8) 23-β-O^{i}Pr	44 ±17.0	16 ±12.0	40 ± 6.0	18 ±11.0
(5) 22-α-Br-23-α,β-OEt	21 ±14.0	28 ±19.0	9.9 ± 9.2	33 ±11.0

* = 20 replications

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References

1. For part 2 see: M.G. Brasca, H.B. Broughton, D. Craig, S.V. Ley, A. Abad. Somovilla and P.L. Toogood, *Tetrahedron Lett.*, 1988, **29**, 1853.
2. a) C.J. Turner, M.S. Tempesta, R.B. Taylor, M.G. Zagorski, J.S. Termini, D.R. Schroeder, and K. Nakanishi, *Tetrahedron*, 1987, **43**, 2789; b) J.N. Bilton, H.B. Broughton, P.S. Jones, S.V. Ley, Z. Lidert, E.D.Morgan, H.S. Rzepa, R.N. Sheppard, A.M.Z. Slawin, and D.J. Williams, *Tetrahedron*, 1987, **43**, 2805; c) W. Kraus, M. Bokel, A. Bruhn, R. Cramer, I. Klaiber, A. Klenk, G. Nagl, H. Pöhnl, H. Sadlo, and B. Vogler, *Tetrahedron*, 1987, **43**, 2817.
3. For recent references, see H. Rembold, and E. de Souza Garcia, *J. Insect Physiol.*, 1984, **30**, 939; A.J. Mordue, P.K. Cottee, and K.A. Evans, *Physiol. Entomol.*, 1985, **10**, 431; K. Feuerhake and H. Schmutterer, *Z. Pfanzenkrankh. Pfanzenschutz*, 1985, **92**, 643; R.B. Yamasaki, and J. A. Klocke, *J. Agric. Food Chem.*, 1987, **35**, 467; I. Kubo, and J.A. Klocke in *Natural Resistence of Plants to Pests, Role of Allelochemicals*, Ed. M.B. Green, and P.A. Hedin, ACS Symoposium series 296, American Chemical Society, 1986, 206.
4. J.N. Bilton, P.S. Jones, S.V. Ley, N.G. Robinson, and R.N. Sheppard, *Tetrahedron Lett.*, 1988, **29**, 1849.
5. S.V. Ley, D. Santafianos, W.M. Blaney, and M.S.J. Simmonds, *Tetrahedron Lett.*, 1987, **28**, 221.
6. W. Kraus, M. Bokel, A. Klenk, H. Pöhnl, *Tetrahedron Lett.*, 1985, **26**, 6435.
7. N.Z. Dimetry, *Experientia*, **32**, 1006.

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