Ferbam were screened against the organism *in vitro*. Poisoned food technique was used with PDA medium. Three doses were tried for each fungicide: (i) dose recommended for field application, (ii) half of the first dose, and (iii) twice the first dose. Radial growth of mycelium was recorded starting from 96 hr of inoculation up to 192 hr. Percentage of growth inhibition was calculated. Benomyl and Bavistin did not significantly inhibit growth of *Alternaria alternata*. Only Ferbam, Dithane M-45 and Captan inhibited the growth of the pathogen by > 50%. Field experiments need to be conducted to determine the effectiveness of these fungicides (Ferbam, Dithane M-45 and Captan).

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STEREOSPECIFIC SYNTHESIS OF OPTICALLY ACTIVE *CIS*-3(R)-(3′,3′-DIHALO-2′-PROPENYL)-2,2-DIMETHYL-1(S)-HYDROXYMETHYL CYCLOPROPANES AND THE CORRESPONDING CYCLOPROPANE-1-CARBOXYLIC ACIDS FROM (+)-3-CARENE

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A number of compounds derived from cyclopropyl methanol/cyclopropane carboxylic acids lacking an asymmetric centre are known to exhibit miticidal activity[1-3]. Staal *et al*[2] attributed the mite ovicidal activity of such compounds to the cyclopropyl methyl moiety. Recently some optically active esters derived from 2,2-dimethyl 3-*n*-propyl cyclopropyl ethanol[4] and cyclopropane acetic acids[5] exhibited miticidal activity against tuber, potato and red spider mites as well as pink and purple mites (adult stage) of tea plantations at dosage levels of 0.0001–1% with 100% mortality. In addition, esters of 2,2-dimethyl 3-alkyl cyclopropane carboxylic acids, optically active[6,7] as well as inactive dl *cis* and *trans* isomers[8] are reported to possess insecticidal activity. With a view to studying the effect of a halogen containing unsaturated alkyl side chain at C_3, in place of the normal saturated alkyl side chain, we now report the synthesis of alcohol moieties (VIII and XIII) as also the corresponding acid moieties (IX and XIV) from (+) 3-carene*. These compounds have been prepared with a view to examining some of the esters prepared from alcohol and acid moieties for evaluating them for miticidal and possible insecticidal and larvicidal activities.

The acetate aldehyde[9] (III), on Wittig reaction using 1,1-dibromomethylene triphenyl phosphorane[10] gave the dibromo acetate (VII), ** $C_{11}H_{16}O_2Br_2$, M^+ 338 (^{79}Br), 340, 342 (^{81}Br), $[\alpha]_D^{28}+4.6°$ (C, 2.0, $CHCl_3$), b.p. 184° (bath)/5 mm; with the following data:

*** IR bands at: 1736 (C=O), 1242 (−OAC), 1630, 790(HC=C<); PMR ($CDCl_3$): Signals at: 0.55 to 0.84(2H, *m*, C_1 and C_3 cyclopropane protons), 0.97, 1.03(3H each, *s* each, gem-dimethyl), 2.01(5H, *s*, overlapping multiplet, acetate methyl and -CH_2 at C_3), 4.04(2H, *t*, J=6.4 Hz, methylene protons of $-CH_2OAc$) and 6.36(1H, *t*, J=6 Hz, olefinic proton).

Alternatively, compound (II) was treated with tribromomethyl carbanion[11] ($CHBr_3/\overset{+}{K}\overset{-}{O}tBu$ at −20°) to afford as a major product (IV, 35%), $C_{11}H_{17}O_2Br_3$, M^+ 418(^{79}Br), 421, 424(^{81}Br), $[\alpha]_D^{27}-15.4°$ (C, 1.96, $CHCl_3$); with the following spectral data: IR bands at: 3400 (−OH), and 1710 (>C=O); PMR (CCl_4) signals at: 0.76–0.9(2H, *m*, C_1 and C_3 cyclopropane protons), 0.96, 1.13(3H each, *s* each, *gem*-dimethyl), 1.76(2H, *m*, $-CH_2$ at C_3), 2.13(3H, *s*, $-COCH_3$), 2.43(2H, *d*, J=7 Hz, $-COCH_2$), and 3.83(2H, *m* $-C_2'-H$ and −OH proton). The other products were not characterized further.

Acetylation (Ac_2O/pyridine) of (IV) gave the acetate (V) as a mixture of diastereomers; $C_{13}H_{19}O_3Br_3$, M^+ 460 (^{79}Br), 463, 466 (^{81}Br), $[\alpha]_D^{27}-5.8°$ (C, 2.0, $CHCl_3$); IR: 1750, 1215 ($-OCOCH_3$), 1710 (>C=O); PMR ($CDCl_3$): 0.62–0.72 (2H, *m*, C_1 and C_3 cyclopropane protons), 0.88, 0.96, 1.0 (6H, *s* each, gem-dimethyls of both diastereomers), 1.86 (2H, *m*, $-CH_2$ at C_3), 2.12, 2.16 (3H each, *s* each, acetate methyls of both diastereomers), 2.40 (2H, *d*, J=6 Hz, methylene adjacent to C=O) and 5.52 (1H, *m*, $-C_2'$ proton of both diastereomers).

Bayer-Villiger oxidation (PBA) of V afforded a mixture of diastereomers of (VI), $C_{13}H_{19}O_4Br_3$, M^+ 476 (^{79}Br), 479, 482 (^{81}Br), $[\alpha]_D^{29}+10.1°$ (C,

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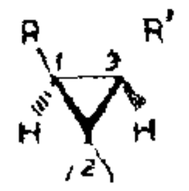
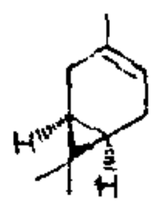
* Optical purity of (+) 3-carene used in the reactions is more than 98%.

** Satisfactory elemental analysis have been obtained for all the compounds reported.

*** IR bands expressed in ν(cm^{-1}) and PMR chemical shifts in δ(ppm) scale with TMS as internal standard.

NCL Communication No. 3998





I

II R = $-CH_2-\overset{O}{\overset{\|}{C}}-CH_3$, R′ = $-CH_2-CHO$

III R = $-CH_2-O-\overset{O}{\overset{\|}{C}}-CH_3$, R′ = $-CH_2-CHO$

IV R = $-CH_2-\overset{O}{\overset{\|}{C}}-CH_3$, R′ = $-CH_2-\underset{CBr_3}{\overset{OH}{CH}}$

V R = $-CH_2-\overset{O}{\overset{\|}{C}}-CH_3$, R′ = $-CH_2-\underset{CBr_3}{\overset{OCOCH_3}{CH}}$

VI R = $-CH_2-O-\overset{O}{\overset{\|}{C}}-CH_3$, R′ = $-CH_2-\underset{CBr_3}{CH}$ ($OCOCH_3$)

VII R = $-CH_2-O-\overset{O}{\overset{\|}{C}}-CH_3$, R′ = $-CH_2-CH=CBr_2$

VIII R = $-CH_2-OH$, R′ = $-CH_2-CH=CBr_2$

IX R = $-COOH$ or $-COOCH_3$, R′ = $-CH_2-CH=CBr_2$

X R = $-CH_2-O-\overset{O}{\overset{\|}{C}}-CH_3$, R′ = $-CH_2-\underset{CCl_3}{\overset{OH}{CH}}$

XI R = $-CH_2-O-\overset{O}{\overset{\|}{C}}-CH_3$, R′ = $-CH_2-\underset{CCl_3}{\overset{OCOCH_3}{CH}}$

XII R = $-CH_2-O-\overset{O}{\overset{\|}{C}}-CH_3$, R′ = $-CH_2-CH=CCl_2$

XIII R = $-CH_2-OH$, R′ = $-CH_2-CH=CCl_2$

XIV R = $-COOH$ or $COOCH_3$, R′ = $-CH_2-CH=CCl_2$

2.0, $CHCl_3$); IR: 1745, 1220 ($-OCOCH_3$); PMR ($CDCl_3$): 0.60–0.70 (2H, *m*, cyclopropane protons at C_1 & C_3), 0.88, 0.90, 0.91 (6H, *s* each, gem-dimethyls of both diastereomers), 1.72, 1.76, 1.82, 1.88 (8H, *s*, overlapping multiplet, acetate methyls of both diastereomers and $-CH_2$ at C_3), 3.5 (2H, *m*, $-CH_2$ at C_1 of both the diastereomers) and 4.68 (1H, *m*, $-C_2'$ proton of both diastereomers).

Treatment of VI with Zn/AcOH in ether at 0°C afforded the same dibromo acetate (VII), (superimposable IR and PMR).

The corresponding dichloroacetate (XII) was prepared from (III) by analogous procedures, via the intermediates (X and XI), $C_{11}H_{16}O_2Cl_2$; M^+ 250 (^{35}Cl), 254 (^{37}Cl); $[\alpha]_D^{30} + 7.7°$ (C, 2.1, $CHCl_3$); IR: 1730, 1230 ($-OCOCH_3$), 1620, 785 (HC=C<); PMR ($CDCl_3$): 0.6–0.9(2H, *m*, C_1 & C_3 cyclopropane protons), 1.06, 1.12(3H each, *s* each, *gem*-dimethyl), 2.08 (3H, *s*, acetate methyl), 2.26(2H, apparent triplet, $-CH_2$ at C_3), 4.14 (2H, *t*, J=6.4 Hz, $-CH_2$ at C_1) and 5.92 (1H, *t*, J=6.4 Hz, olefinic proton).

Saponification (KOH/MeOH) of acetates (VII) and (XII) afforded the alcohols (VIII) and (XIII) respectively.

Compound (VIII), $C_9H_{14}OBr_2$, M^+, 296 (^{79}Br), 298, 300 (^{81}Br), $[\alpha]_D^{28} - 0.54°$ (C, 2.2, ($CHCl_3$), b.p. 161° (bath)/8 mm; IR: 3320 (−OH), 1620, 790 (HC=C<); PMR ((($CDCl_3$): 0.6 – 0.94(2H, *m*, C_1 & C_3 cyclopropane protons), 1.1, 1.16(3H each, *s* each, gem-dimethyl), 1.66(1H, *s*, −OH proton), 2.16(2H, apparent triplet, $-CH_2$ at C_3), 3.7 (2H, *d*, J=6.4 Hz, $-CH_2$ at C_1) and 6.48 (1H, *t*, J=6.4 Hz, olefinic proton).

Compound (XIII), $C_9H_{14}OCl_2$, M^+, 208 (^{35}Cl), 212 (^{37}Cl), $[\alpha]_D^{29} + 4.05°$ (C, 2.0, $CHCl_3$), b.p. 164° (bath)/3 mm, IR: 3310 (−OH), 1620, 780 (HC=C<); PMR ($CDCl_3$): 0.76–0.92(2H, *m*, C_1 & C_3 cyclopropane protons), 1.04, 1.08(3H each, *s* each, gem-dimethyl), 1.62(1H, *s*, −OH proton), 2.22(2H, apparent triplet, $-CH_2$ at C_3), 3.72 (2H, *d*, J=6.4 Hz, hydroxy methyl at C_1) and 5.94 (1H, *t*, J=6.4 Hz, olefinic proton).

Jones chromic acid oxidation of (VIII) and (XIII) followed by esterification (diazomethane) gave (IX) and (XIV) respectively.

Ester (IX), $C_{10}H_{14}O_2Br_2$, M^+, 324 (^{79}Br), 326, 328 (^{81}Br), $[\alpha]_D^{28} + 6.0°$ (C, 1.5, $CHCl_3$), b.p. 167° (bath)/4 mm, IR: 1730 (>C=O), 1620 (C=C); PMR ($CDCl_3$): 0.86(1H, *m*, C_3 cyclopropane proton), 1.18, 1.22(3H each, *s* each, gem-dimethyl), 1.5(1H, *d*, J=6 Hz, C_1 cyclopropane proton), 2.5 (2H, *m*, $-CH_2$ at C_3), 3.68(3H, *s*, ester methyl) and 6.38(1H, *t*, J=6 Hz, olefinic proton).

Ester (XIV),$C_{10}H_{14}O_2Cl_2$, M^+, 236 (^{35}Cl), 240 (^{37}Cl), $[\alpha]_D^{25} - 2.84°$ (C, 2.6, $CHCl_3$), b.p. 168° (bath)/5 mm, IR: 1730 (ester C=O), 1620, 780 (HC=C<); PMR ($CDCl_3$): 0.84 – 1.1(1H, *m*, C_3 cyclopropane proton), 1.12, 1.16(3H each, *s*, each, gem-dimethyl), 1.52(1H, *d*, J=6 Hz, C_1 cyclopropane proton), 2.48(2H, *m*, $-CH_2$ at C_3), 3.68(3H, *s*, ester methyl) and 5.84(1H, *t*, J=6.4 Hz, olefinic proton).

30 August 1986; Revised 11 December 1986

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A SIMPLE AND INEXPENSIVE METHOD OF MASS SCREENING FOR ABNORMAL HAEMOGLOBINS

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ABNORMAL haemoglobins are detected by electrophoresis in different media and buffer systems[1–4]. Electrophoresis on cellulose acetate, although extensively used, is expensive and thus limits mass survey of carrier traits for haemoglobinopathies. A rapid, simple and relatively inexpensive method for haemoglobin electrophoresis is described here.

Red cell haemolysate (10g/dl) was prepared by the standard method[1]. 1% Agarose (Gibco Laboratories, USA) in tris-EDTA-borate(TEB) buffer, pH 8.9 (tris-hydroxymethyl aminoethane 14.4 g; EDTA 1.5 g; boric acid 0.9 g; and water to 1 l) was prepared by heating until the solution becomes clear. This solution (18 ml) was poured on a glass plate (17 cm × 8 cm × 0.1 cm) fitted to a perspex frame of 1–2 mm thickness and covered by a glass plate of the same size and allowed to set for 30 min. The covering plate and the frame are removed and the plate with gel side up is placed in an electrophoresis trough (Shandon, UK). The buffer tanks are filled with TEB buffer pH 8.9 and contact is established with filter paper strips and a constant current of 24 m Amp at 150V was passed for 15 min (Vokam stabilized DC power supply, Shandon, UK). Haemolysates are applied to the gel plate using a multi-applicator (Shandon, UK) and the same current was passed for 60–90 min. The plate was removed, fixed in 5% acetic acid in 85% ethyl alcohol for 15 min, stained with Nigrosine Black (0.5 Nigrosine Black in 1% acetic acid) for 10 min and dried overnight on a filter paper. A permanent record may be kept by a photograph (figure 1) or a photocopy.

Figure 1 shows the preparation of haemoglobins on agarose gel plate. Samples 1,6,9 and 16 are from normal control, samples 2,7, 11 and 15 from patients E B-thalassaemia showing haemoglobin E and B bands, samples 3,8,10 and 12 are from haemoglobin

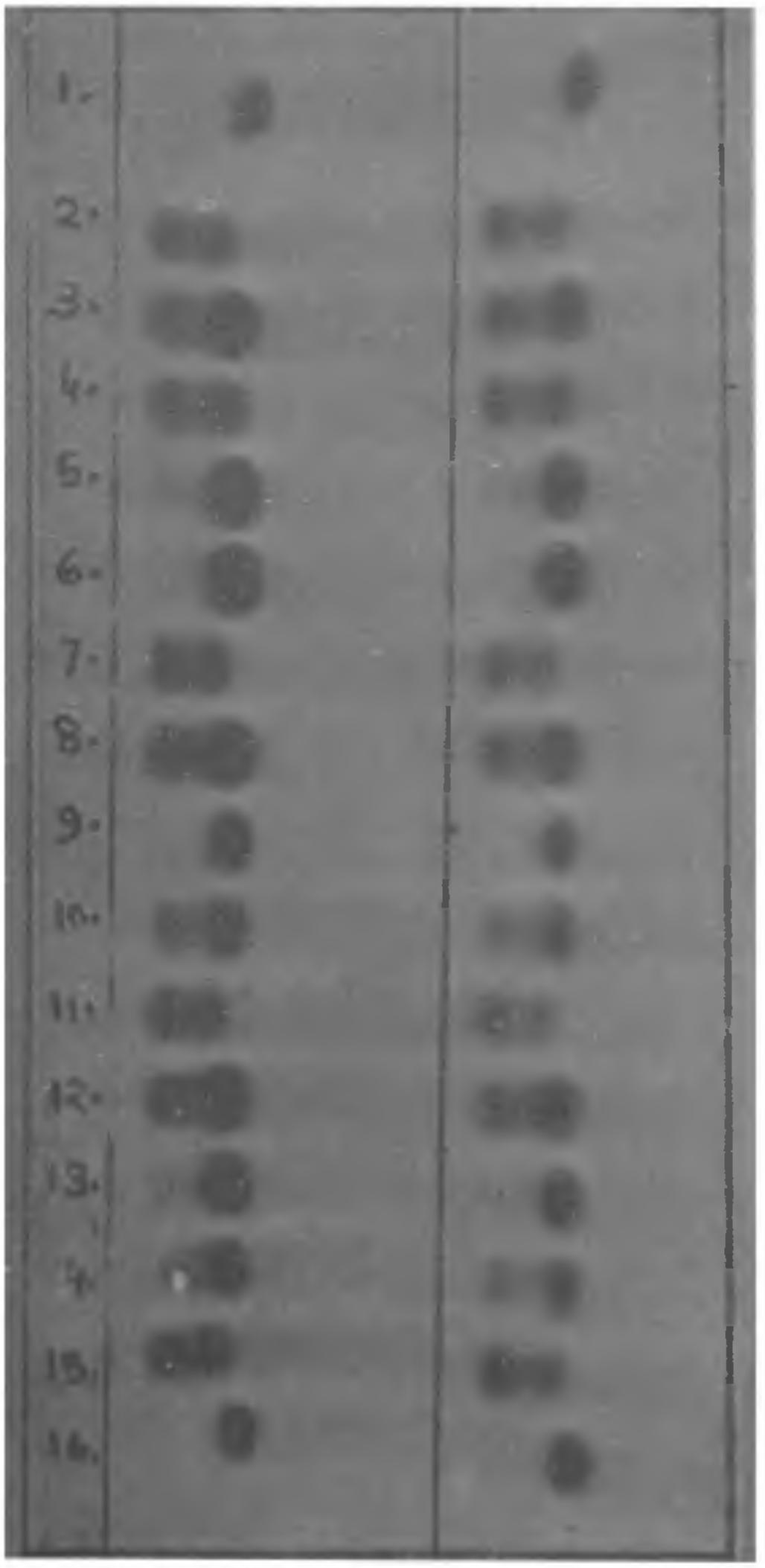

Figure 1. Agarose gel plate electrophoresis showing normal haemoglobin, beta-thalassaemia carrier state, haemoglobin E carrier state and E-Beta thalassaemia.