pound ketone and anisic acid, which were identified by a comparison (m. m.p., TLC, IR and NMR) with authentic samples.

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ABSCISIC ACID IN *HYACINTHUS ORIENTALIS* BULBS

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Key Word Index—*Hyacinthus orientalis*; Liliaceae; hyacinth; abscisic acid.

INTRODUCTION

The plant growth hormone abscisic acid (ABA) has been detected in many different plants.[1–3] Hyacinth bulbs (*Hyacinthus orientalis* L.) require a period of high temperature treatment (20°) for flower bud development, then a period of low temperature treatment, a few degrees above the freezing point, for dormancy release and flower growth.[4,5] Consideration of hormonal control of the dormancy state in a plant[6–8] requires the identification of abscisic acid, and this was undertaken.

RESULTS AND DISCUSSION

The presence of an acidic inhibitor in dry and fleshy scales of hyacinth bulbs was found by PC and growth inhibition of wheat coleoptiles. This showed a potent inhibitory substance at R_f corresponding to those of synthetic ABA. Further purification of the inhibitory fraction by TLC and GLC of the methylated derivative of this inhibitor on two different columns identified it as abscisic acid.

Abscisic acid was identified in dry as well as in fleshy scales of hyacinth bulb.

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EXPERIMENTAL

Plant material. Hyacinth bulbs (cv. Lady Derby) were collected in June. The dry blue scales were removed and these and the remainder of the bulbs were analyzed.

Extraction. The bulbs without scales (1·5 kg) and scales (0·5 kg) were homogenized in 80% EtOH (1 kg fr. wt to 4 l.). The extract was centrifuged and the residue was discarded, the EtOH was evaporated under vacuum at 40°, and the ether-soluble acid fraction prepared.

PC and TLC. The acid fraction was chromatographed on Whatman 3 paper ($PrOH–NH_4OH–H_2O$, 10:1:1) using synthetic ABA (Hoffman La-Roche) as a standard. The wheat coleoptile bioassay[9] revealed the presence of an inhibitory band at R_f corresponding to that of synthetic ABA. The material between R_fs 0·60–0·80 was eluted with MeOH and chromatographed on TLC plates of silica gel GF_{254} ($C_6H_6–CHCl_3–HCOOH$, 12:8:1) with synthetic ABA as a standard. The zone of quenched fluorescence adjacent to the ABA marker was eluted. An aliquot was tested for growth-inhibitory activity, and the residue was chromatographed with $C_6H_6–Me_2CO–HOAc$ (70:30:1). Two fluorescence-quenching bands appeared by the standard ABA, the one at higher R_f was extracted and esterified with diazomethane in Et_2O.

GLC. The GLC parameters were as follows (using a Barber Coleman gas chromatograph with an electron capture detector): Column 1: 5·1% QF-1 on 60/80 mesh GasChrom Q, 0·6 × 244 cm, Column 2: 2·6% SE-30 on 60/80 mesh GasChrom Q, 0·6 × 183 cm. Columns temp. 187° isothermal, injector temp. −213°, carrier gas N_2, 75 ml per min. The methylated compound has T_R 14·6 min on Column 1 and T_R 4·3 min on Column 2. (Std. ABA T_R 14·5 min, Column 1, and T_R 4·3 min, Column 2).

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