J. Pharm. Pharmacol., 1965, **17**, *Suppl.*, 41S–43S Received May 3, 1965

Carvone in the developing fruits of *Anethum graveolens* L. and *Carum carvi* L.

T. J. BETTS

The carvone content of the developing fruits of *Anethum graveolens* L. (dill) and *Carum carvi* L. (caraway) has been determined spectrophotometrically over three seasons. Three or four weeks after pollination, carvone in the fruits attains a level of 11–20 mg per 100 caraway fruits, or 4–9 mg per 100 dill fruits. Variations in the carvone content of the essential oils distilled from these fruits are probably due to variable quantities of limonene.

THE principal flavouring constituent of the umbelliferous fruits *Anethum graveolens* L. (dill) and *Carum carvi* L. (caraway), (+)-carvone, forms up to 63% of the essential oils of these two fruits (B.P.C.). The other main constituent, (+)-limonene, is probably the direct biogenetic precursor of (+)-carvone (Birch, 1963). The oils have similar physical constants and may be indistinguishable, although dill oil may contain small amounts of aromatic compounds such as dillapiole (Gupta, Chandra & Zaidi, 1955). The present communication examines the production of carvone in the fruits of these closely related species.

Experimental

Plant material. Caraway and dill plants, grown in the Myddelton House drug garden, Enfield, Middlesex, were identified by their botanical characters, the macroscopy of their fruits and the constituents of their essential oils. Compound umbels were marked when most of their flowers were open and insect pollination was taking place. Fruits were then sampled at various times after being marked, until they ripened and dropped. Samples were taken in this fashion during 1962, 1963 and 1964.

Distillate extracts. Each sample was of the same population of 100–200 accurately counted fresh entire cremocarps (fruits) taken from about five compound umbels on different plants. At the final, ripe stage, two mericarps were counted for each cremocarp. Carpophores and small amounts of pedicel were included with the cut off fruits, together, during the first week after pollination, with any petals remaining. Only healthy plants and full sized fruits were sampled.

The fruits were weighed and steam distilled the same day by co-distilling with about 50 ml water until 5 ml liquid remained in the still; the aqueous condensate was passed directly into a separating funnel containing 4 ml n-hexane. When the distillation was completed (40–60 min), the condenser was washed out into the separating funnel with 1 ml n-hexane and a little water. The funnel contents were shaken gently and

From the Department of Pharmacognosy, School of Pharmacy, University of London, Brunswick Square, London, W.C.1.

allowed to separate and the separated hexane layer diluted to volume with hexane in a volumetric flask. In the first weeks of fruit development this volume was 5 ml; in later weeks, 10 or 20 ml, this being made up by re-extracting the aqueous condensate with more hexane.

In both 1962 and 1963, two identical samples were taken, the second being used to determine the loss of weight on heating at 105°. This value was subsequently corrected for the essential oil content of the sample to provide a value for the dry weight of each sample (fresh weight minus water only). The total oil content was taken as twice that of the carvone found.

Thin-layer chromatography of the above hexane extracts was carried out as previously described (Betts, 1964).

Carvone assay. A modification of the method of Stenlake & Williams (1957) was used. A suitable aliquot of the hexane extract of the distillate (0·5–2 ml) was diluted to 10 ml with n-hexane, absolute ethanol (5 ml) was added, and the solution was refluxed (4½ hr) with glacial acetic acid (0·5 ml) and Girard's Reagent T (200 mg). After cooling, N sodium hydroxide (7·5 ml) was added and the mixture was transferred to a separating funnel with water and ether. The organic phase was extracted three times with water, each extract being washed with ether. The combined aqueous extracts were diluted with water to a suitable volume (200–500 ml) and the absorbance determined at approximately 273 mμ. A blank assay was made with 10 ml n-hexane and used as the reference solution. The amount of carvone present in the sample was calculated as follows:

$$\text{Carvone, mg per 100 fruits} = \frac{d \times v}{1{\cdot}267 \times n}$$

Where d = absorbance reading at peak; v = volume (ml) of diluted aqueous extract prepared for reading d; n = number of fruits represented by v (i.e., by the hexane aliquot taken for assay).

The high extinction value of the water-soluble carvone complex [E(1%, 1 cm) 1267] means that the assay is sensitive enough to work with about fifteen developed fruits.

Results and discussion

Similar results were obtained for carvone assays on the developing fruits over three seasons, and illustrative figures are quoted in Table 1. At 1 week after pollination, caraway fruits contained little or no carvone, but by the third week 10 mg or more per 100 fruits was present. In subsequent weeks the carvone content varied between 12–20 mg per 100 fruits in 1962 and 11–14 mg in 1964. Dill fruits initially contained more carvone (1–2 mg per 100 fruits) than caraway fruits but about 3 weeks after pollination the carvone content was only 5–9 mg per 100 fruits in 1962 and 4–6 mg in 1964. These results support Luyendijk's observation (1957) that as the two fruits develop "the content of carbonyl compounds increases from traces until the specific level for the species is reached".

The distinction between carvone content expressed in terms of dry weight and as content of 100 fruits arises because the carvone content rises more rapidly than does the dry weight. Thin-layer chromatography confirmed that carvone was present at all stages of the development of both dill and caraway fruits. Dillapiole was not observed. From spot size and intensity, the terpene hydrocarbons (e.g. limonene) decrease in quantity as the fruits of caraway and dill develop and ripen.

TABLE 1. OBSERVATIONS ON DEVELOPING FRUITS 1963

Date marked i.e. in flower	Date collected	Days (and weeks) since marking	Fresh wt. 100 fruits[1] in mg	% fresh wt. not lost on drying	Calc.[2] dry wt. 100 fruits in mg	mg carvone	
						in 100 fruits[3]	per g dry wt.
			Caraway				
31 May	7 June	7 (*1*)	426	19	82	0·4	5
7 June	21 June	14 (*2*)	546	21	127	6	47
31 May	21 June	21 (*3*)	1,160	24	311	15	47
7 June	5 July	28 (*4*)	1,324	29	412	12	30
31 May	5 July	35 (*5*)	1,391	35	515	16	31
31 May	12 July	42 (*6*)	904	60	575	16	30
			Dill				
6 Aug.	15 Aug.	9 (*1*)	268	26	75	2	24
29 July	15 Aug.	17 (*2*)	701	23	174	6	33
6 Aug.	28 Aug.	22 (*3*)	838	29	257	7	28
29 July	28 Aug.	30 (*4*)	1,062	33	357	6	17
6 Aug.	11 Sept.	36 (*5*)	1,154	33	399	7	17
29 July	11 Sept.	42 (*6*)	513	58	313	8	26

[1] Fresh weights are averages of 2 or 3 lots.
[2] Dry weights obtained from percentage of fresh weight not lost on drying, corrected for loss of essential oil (taken to be twice the amount of carvone present).
[3] Average of 2–4 determinations.

Although the maximum yield of carvone can be obtained from a crop of fruits which has been allowed to develop for about 4 weeks, the essential oil from this will contain a higher proportion of limonene than that from more developed fruits and so will be sweeter and less intensely flavoured. Low carvone content of the essential oil is thus not necessarily the result of carvone deficiency in the fruit.

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