TABLE II

	Stigma and Glumes coloured	Stigma coloured, glumes colourless	Stigma colourless, glumes coloured	Stigma and glumes colourless	X^2	P
Obs. ..	281	64	55	154	..	..
(i) Cal. (162 : 94 × 162 : 94) Ind. (Ghose)	221·8	123·7	128·7	74·8	174·123	Very small
(ii) Cal. (2·02% C O.) (D'Cruz)	291·9	58·7	58·7	144·7	2·143	0·30–50
(iii) Cal. (504 : 117 : 108 : 295) (New)	272·7	63·3	58·4	159·6	0·6546	80–90

The inhibitor differentially modifies the interaction. There is no linkage.

A critical study of literature reveals that many anthocyanin genes have pleiotropic effects affecting from 2 to 11 characters. Shrivastava *et al.* (1968) report monogenic inheritance of colour in four parts, leaf-sheath, ligule, stigma and apiculus as a group in a variety JBS-294, which developed pigment in 11 parts. The results obtained by these authors, however, are better explained on the basis of 189 : 67 ratio, that is four colour genes. Recently Kadam (1970) has shown, in a reanalysis of Mitra and Ganguli's data (1932) in a wild rice of Assam that 4 colour genes cause development of anthocyanin pigment in 11 characters by teaming up in a variety of ways and an inhibitor affects two of the eleven characters.

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PRELIMINARY NOTE ON THE EFFECTS OF INCUBATION OF FRESH TUBERS OF DIOSCOREA ON DIOSGENIN CONTENT*

SEVERAL attempts to increase the diosgenin content in Dioscorea tubers by incubation of freshly harvested tubers have been reported in recent years. The results have been variable depending upon the species and specific conditions for incubation. For example, the diosgenin content of *D. floribunda* was found to have increased when the incubation took place in the presence of squalene—a precursor of diosgenin biosynthesis, in the dark at 37° C[1]. However, no increase was observed in *D. belizensis* at 25° C and 37° C with 5 days' incubation time[2]. In another study it was observed that the apparent increase in diosgenin content of *D. belizensis* tubers incubated for 21 days was due to loss of dry matter and was not a real increase[3]. In order to further study the effects of incubation on diosgenin content, experiments were conducted with two diosgenin containing species, *viz.*, *D. floribunda* and *D. deltoidea* which are growing at the Hesaraghatta Farm of the Indian Institute of Horticultural Research, Bangalore. The results of these experiments are reported in this paper.

D. floribunda and *D. deltoidea* were selected for this study as these two species have been found to be more promising in the continuing programme on development of Dioscorea as a cultivated crop for diosgenin production. Tubers from one year old clones of the two species were used in these studies. Fresh tubers after cleaning with distilled water and removing the adhering water were powdered in an iron mortar and pestle. Forty grams of powdered tuber was taken for each treatment and was homogenised with 200 ml of distilled water for 5 minutes in a blender. Moisture content was determined on two samples weighed out of the original powdered material from the tuber. The homogenised slurry was incubated for 0, 6, 12, 24, 48 and 72 hours at 37° C. At the end of the requisite time of incubation, concentrated hydrochloric acid was added. The final concentration of HCl in the slurry was 2·5 N in each treatment. The sample after hydrolysis for 2 hours was assayed for diosgenin according to the procedure described earlier[4].

The results obtained with *D. deltoidea* are given in Table I. There was an increase in diosgenin content of 0·24 gm and 0·39 gm per 100 gm of dry matter after 12 and 48 hours of incubation time. However, this increase as well as the marginal decrease observed at 6, 24 and 72 hours of incubation were statistically insignificant.

TABLE I

Effect of incubation on diosgenin content in D. deltoidea

Incubation time	*Dioscorea deltoidea* — Percentage of diosgenin		Increase or decrease over control
	On fresh weight basis	On dry weight basis	
Control (0 hours) ..	1·25	3·45	..
6 ,, ..	1·24	3·44	−0·01
12 ,, ..	1·33	3·69	+0·24
24 ,, ..	1·24	3·44	−0·01
48 ,, ..	1·39	3·84	+0·39
72 ,, ..	1·22	3·38	−0·07

Average of two analyses. Moisture per cent 63·96.
Not significant.

The results obtained with *D. floribunda* are given in Table II. There was an increase of

TABLE II

Effect of incubation on diosgenin content in D. floribunda

Incubation time	*Dioscorea floribunda* — Percentage of diosgenin		Increase or decrease over control
	On fresh weight basis	On dry weight basis	
Control (0 hours) ..	1·27	3·80	..
6 ,, ..	1·42	4·24	+0·44
12 ,, ..	1·48	4·42	+0·62
24 ,, ..	1·48	4·42	+0·62
48 ,, ..	1·30	3·88	+0·08
72 ,, ..	1·06	3·15	−0·65

Average of two analyses. Moisture per cent 66·43.
C.D. at 5% = 0·386.

0·44 (11·5%), 0·62 (16·3%), 0·62 (16·3%) and 0·08 (2·1%) grams of diosgenin over the control after 6, 12, 24 and 48 hours of incubation respectively. However, the diosgenin content showed a decrease of 0·65 gm for 100 gm of dry matter after 72 hours incubation. The observed increase after 6, 12 and 24 hours and the decrease observed in 72 hours treatment were statistically significant. The observed increase after incubation is real as the dry matter was kept constant in all treatments.

Quantitative estimation[5] of squalene in fresh tubers revealed considerable differences. The content was 7·97 mg/100 gm in *D. floribunda* and 2·66 mg/100 gm of fresh tubers in *D. deltoidea*.

The behaviour of the two species is quite distinct from each other. There was no significant increase or decrease in *D. deltoidea* for upto 72 hours of incubation. On the other hand *D. floribunda showed* an increase for upto 24 hours. Beyond 24 hours there was trend towards decrease.

Similar increases as observed here in *D. floribunda* have been attributed to be due to enhanced enzymatic activity on incubation in *Balanites orbicularis*[6]. The increase observed in *D. floribunda* could also be due to the activity of certain micro-organisms associated with tubers which may be converting certain procursors of diosgenin to diosgenin.

Micro-organisms associated with tubers could also be responsible for the decreases observed during incubation. It has been reported[7] that certain micro-organisms, particularly some fungi, have the ability to bind steroids on prolonged incubation thus making them more difficult to extract by conventional methods. The loss in diosgenin content could also be due to degradation of sapogenin. The degradation upon prolonged fermentation under non-sterile conditions at 37° C has also been reported during microbial hydrolysis of Dioscorea[8].

The differences in squalene content could also be responsible for differential response of the two species to incubation.

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