

TABLE I

The  $R_f$  values and absorption maxima of the converted anthocyanidin and pure cyanidin chloride

Genotype	$R_f$ values		Absorption maxima in $m\mu$	
	Baw	Forestal	UV	Visible
$a_2$	0.68	0.48	279	541
in $a_2$	0.68	0.47	278	542
Cyanidin chloride	0.69	0.49	279	543

anthocyanin. In the known gene action sequence, there are only three genes  $A_2$ ,  $Bz_1$  and  $Bz_2$ , which may convert leucocyanidin to anthocyanin. The  $A_2/a_2$  gene could act in two different ways either by direct conversion of leucocyanidin to cyanidin or on a common precursor in the biosynthesis of these pigments. The glycosidation of quercetin to its 3-glycoside (isoquercetin) by an enzyme from  $Bz_1$  pollen extracts suggests indirectly that this gene might control the glycosidation of anthocyanin molecule.<sup>9</sup> Further studies on

isolation and characterisation of gene-controlled intermediates in anthocyanin biosynthesis, in maize, may unravel the mechanism of gene action at chemical level.

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## IN VITRO GROWTH REQUIREMENTS OF MATURE ENDOSPERM OF *RICINUS COMMUNIS*

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SATSANGI and Mohan Ram (1965) succeeded in inducing embryoid formation in the callus of mature endosperm of *Ricinus communis*, but failed to obtain organogenesis. The present investigation was undertaken (a) to ascertain the growth requirements, and (b) to explore the possibility of induction of root and shoot.

Ripe fruits were collected from the Delhi University Campus. After removal of the fruit-wall and seedcoat, the mature endosperm (with embryo intact) was washed with 'cetavlon' (cetrimide concentrate, diluted to 100 times) and surface-sterilized with chlorine water for 10-12 min. The endosperm was then washed with sterile distilled water and soaked in it for 24 hr. This was then implanted aseptically on modified White's semi-solid (agar 0.8%) medium containing 2% sucrose (WB). The supplements used were: indoleacetic acid (IAA), indolebutyric acid (IBA), indolepropionic acid (IPA), naphthaleneacetic acid (NAA), phenoxyacetic acid (PAA), 2,4-dichlorophenoxy acetic acid (2,4-D), 2,4,5-trichlorophenoxyacetic acid (2,4,5-T), adenine (AD), adenine sulphate (ADSO<sub>4</sub>), kinetin (KN), benzyladenine (BA), diphenyl urea

(DPU), 6-( $\gamma$ , $\gamma$ -dimethylallylamino)-purine (6- $\gamma$ , $\gamma$ ), SD 8339, triacanthine (TA), zeatin (ZN), myo-inositol, casein hydrolysate (CH), coconut milk (CM), and yeast extract (YE). The effect of various sugars was also tested.

On WB alone, WB + IAA, WB + IBA, WB + IPA, WB + NAA, or WB + KN, after six weeks, in 80% cultures, the embryo developed into a normal seedling (Fig. 1, A). However, on WB + 2, 4-D, germination of the embryo was delayed and swelling of the radicle was observed after 3 weeks. The endosperm showed only slight swelling and cracks but did not callus. On WB + 2, 4-D (2 ppm) + KN (5 ppm) + YE (2,500 ppm), germination of the embryo was altogether suppressed, and the radicle callused. Along the surface in contact with the embryo, the endosperm also proliferated forming a white, fluffy callus (Fig. 1, B). In subcultures maintained on the same medium, the endosperm callus showed satisfactory growth and became compact after four passages (each passage of four weeks) (Fig. 1, C).

Squash preparations of a 4-week-old endosperm callus revealed thin-walled cells of different shape and size, while a 10-week-old



callus showed tracheid-like cells with reticulate thickenings. The sections ( $12\text{--}15\ \mu$  thick) of a 12-week-old compact callus exhibited a peripheral zone of cambium-like cells followed by thick-walled cells simulating vascular bundles (Fig. 1, D).

In one set of experiments different concentrations of sucrose ( $0.3 \times 10^{-1}\text{ M}$  to  $3.5 \times 10^{-1}\text{ M}$ ) were used, and fresh and dry weight

recorded after six weeks.  $10^{-1}\text{ M}$  produced best growth of callus.

In another set of experiments, sucrose was replaced by glucose, fructose, maltose, raffinose, arabinose, sorbose, galactose, lactose, mannose and cellobiose (all at equimolar concentration of  $10^{-1}\text{ M}$ ). Sucrose, glucose, fructose and maltose were almost equally effective in bringing about both fresh and dry weight

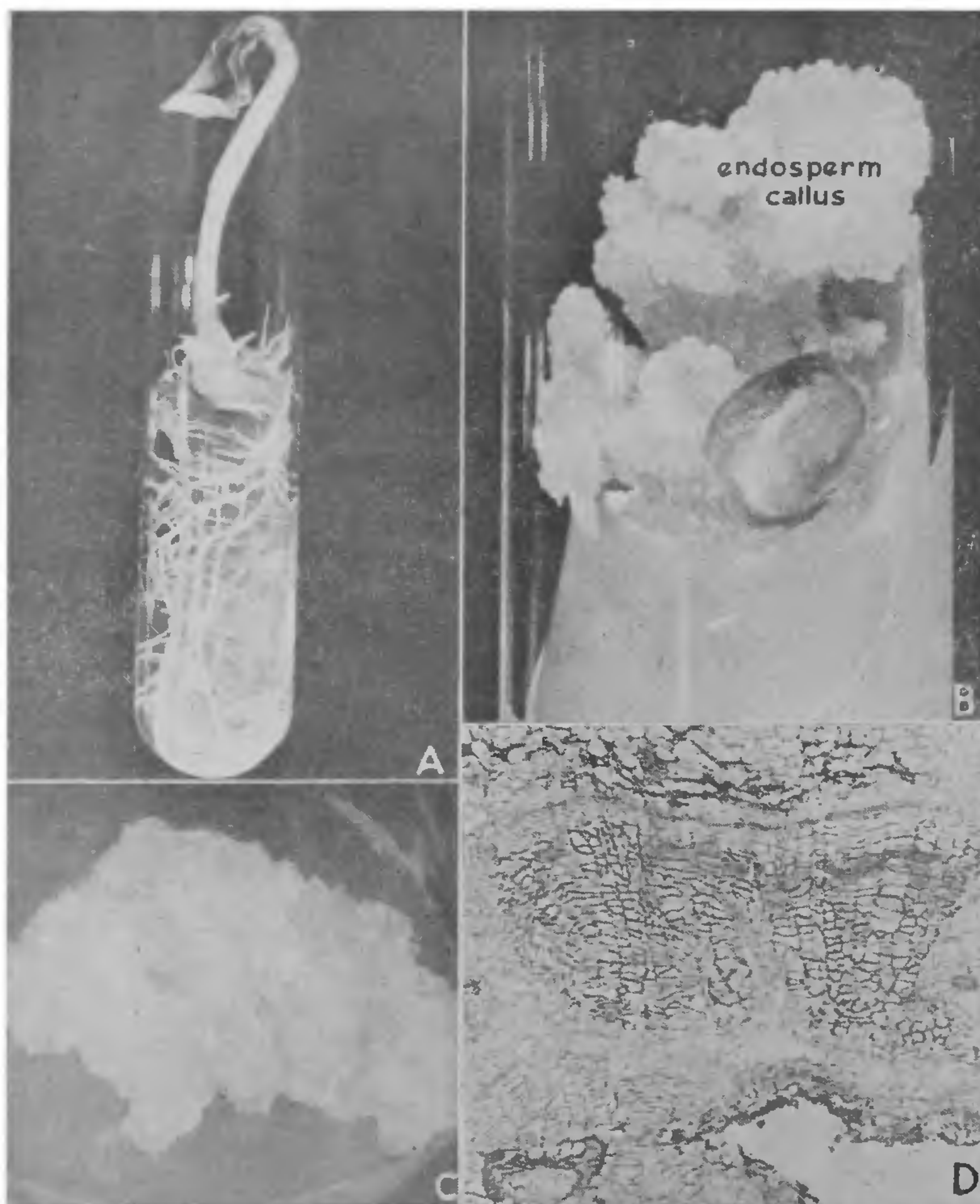


FIG. 1, A-D. A. 6 week old normal seedling on WB,  $\times 1$ ; B. Callused endosperm and collapsed cotyledons in a 8-week-old culture on WB + 2, 4-D + KN + YE,  $\times 2.5$ ; C. Compact endosperm callus (medium same as for B,  $\times 2$ ). D. Vascular bundle-like structure,  $\times 130$ .



increases of callus. These results are summarized in Table I.

TABLE I  
Growth of callus on WB with different supplements

Medium	Concentration	% Increase	
		Fresh weight*	Dry weight*
WB+ sucrose (control) ..	$10^{-1}$ M	1,360	375
WB+ mannose ..	$10^{-1}$ M	145	28
WB+ 2, 4-D ..	$10^{-5}$ M	1,360	430
WB+ 2, 4, 5-T ..	$10^{-5}$ M	695	185
WB+ SD 8339 ..	$2.5 \times 10^{-5}$ M	1,425	470
WB+ DPU ..	$2.5 \times 10^{-5}$ M	470	140
WB+ YE ..	3,000 ppm	1,425	385

\* Average of 48 cultures; growth period 6 weeks.

Of PAA, 2, 4-D and 2, 4, 5-T, at equimolar concentration of  $10^{-1}$  M, 2, 4-D was most effective.

Of SD 8339, KN, BA, 6- $\gamma$ ,  $\gamma$ ; ZN, AD, TA and DPU, optimal increase of fresh and dry weight occurred with SD 8339, KN and ZN. DPU was least effective.

Recently, the author isolated a white, fragile nodulated callus from the endosperm tissue grown on WB + 2, 4-D + KN + YE. This has been successfully subcultured on WB + CH (1,000 ppm).

Thus, the mature endosperm tissue of *Ricinus communis* can be stimulated to divide and form a continuously-growing callus. All efforts to induce organogenesis in callused endosperm have not yet succeeded, and further studies in this direction are in progress.

I am indebted to Professor B. M. Johri for guidance.

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## THE SYSTEMATIC POSITION AND OCCURRENCE OF *PARIOGLOSSUS* (TELEOSTEI : GOBIOIDEA) IN INDIAN WATERS

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THE marine gobioid genus *Parioglossus* Regan 1912<sup>1</sup> (syn. *Herrea* Smith, 1931,<sup>2</sup> not *Herrea* Whitley, 1930,<sup>3</sup> replaced by *Herreolus* Smith, 1931<sup>4</sup>) is of widespread Indo-West Pacific distribution, and comprises four tiny species, the type *P. taeniatus* Regan 1912, originally described from Aldabra,<sup>1</sup> *P. rainfordi* McCulloch, 1921 (Bowen, Queensland<sup>5</sup>), *P. borneensis* Koumans, 1953 (Balikpapan, Borneo<sup>6</sup>), and *P. dotui* Tomiyama, 1958 (SW Japan<sup>7</sup>), the last reaching the maximum size of 37 mm<sup>8</sup> recorded for the genus. *Parioglossus* has been reviewed by Tomiyama,<sup>9</sup> who provides a key and illustrations of these species.

Because of its separate pelvic fins, *Parioglossus* has been placed in the family Eleotridae of the suborder Gobioidea (Regan,<sup>10</sup> Koumans<sup>6</sup>), whose largest family, the Gobiidae, is traditionally characterised by the possession of a shallow ventral disc formed by fusion of the pelvic fins. Although a classification of gobioid fishes based on their osteology was proposed many years ago by Regan,<sup>10</sup> and has been subsequently amplified (Gosline,<sup>11</sup> Hoese<sup>12</sup>), most representatives of the suborder await comprehensive skeletal investigation and, consequently, in several important monographs, have been arranged by external features (Herre,<sup>13</sup> Koumans,<sup>6-14</sup> Smith,<sup>15</sup> etc.).

Recently, the present author has examined alizarin preparations (Fig. 1) and radiographs of the skeleton in *Parioglossus*. While certainly gobioid, with unossified suspensorial foramen (bounded by symplectic, quadrate, and preopercular), and upper (epaxial) and lower (hypaxial) caudal radials, the genus was found however to be gobiid rather than eleotrid in structure. Typical gobiid features displayed are the single pterygoid element in the palatoquadrate arch, markedly T-shaped palatine head, reduced hypercoracoid, five branchiostegous rays, and only one epural plate in the caudal skeleton. *Parioglossus* may thus be regarded as a gobiid specialised externally by secondary separation of the pelvic disc (a trend recorded in many others of the same family<sup>16</sup>) and with further modifications, such as incipient duplication of interspinous bones between neural and haemal spines, forward shifting of the jaw articulation by suspensorial elongation, etc., probably connected with its nektonic mode of life.<sup>17-18</sup> In all the latter features, *Parioglossus* shows affinity with *Ptereleotris* Gill, 1863, *Oxymetopon* Bleeker, 1861, *Ioglossus* Bean, 1882, and probably other genera, all larger free-swimming or burrow-dwelling gobiids with divided pelvic fins.<sup>12-18 19</sup> Another clue to relationships is afforded by the