roves are *Phoenix peludosa* Roxb., *Suaeda maritima* Dumort and *Sonneratia apetala* Ham. salinity 32‰ S.

3. Habitat – c: Intertidal mudflat at Prentice Island (21°43 to 21°46 N and 88°18 to 88°19 E), abundant mangrove vegetations are *Avicennia officinalis* L. *Avicennia marina* Vierh and *Ceriops decandra* (Grifi) Ding hon. salinity 28‰ S.

Soil samples around roots of different mangroves were collected and processed by following modified Baermann funnel method using tapwater. After extraction, the specimens were fixed in hot 4 : 1 F.A. and then mounted on anhydrous glycerine after proper dehydration in glycero-alcohol. The pH and salinity of the interstitial water were determined by standard methods. The stylet-bearing nematodes parasitizing mangrove plants comprise both dorylaimid and tylenchid groups (table 1). Dominant families were represented by Leptonchidae and Dorylaimidae of the order Dorylaimida and Tylenehidae, Anguinidae and Criconematidae of the order Tylenchida. The nematodes under consideration displayed habitat preference and some were strictly restricted to one habitat type. The genera like *Laimydorus*, *Doryllium*, *Nygolaimus*, *Hirschmannialla* and *Helichotylenchus*, etc. exhibit regional abundances only in the intertidal zones of Gangasagar mangrove swamp (habitat – b), Sagar Island and the genera like *Indoditylenchus*[5], *Proleptonchus* and *Tylenchus* were displayed in the intertidal mudflat of Prentice Island (habitat – c). On the other hand, the genera like *Timmus*, *Dorylaimoides*, *Thonus*, *Paralongidorus*, *Paraoxydirus*, *Nygolaimoides*, *Nothocriconema* and *Hemicriconemoides* were predominant in the mudflat of Harinbari mangrove swamp (habitat – b). It is to be noted that all these nematode species were recovered from midlittoral zones of different intertidal habitats in the upper 0–5 cm of the core sample.

Since there are no records of stylet-bearing nematodes parasitizing mangrove flora of the Indian part of the Sundarbans delta, all the nematodes communicated in this paper are supposed to be the first record from India. However, a new host plant *Excoecaria agallocha* has been recorded for *Hemicriconemoides sundarbanensis*[4].

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ENDOSPERM AMYLASE ACTIVITY IN PEARL MILLET SEEDLINGS IN RELATION TO DWARFISM

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It is now well-established that GA_3 stimulates the synthesis and release of α-amylase[1], ribonuclease and protease[2]. This GA has been reported to induce *de novo* synthesis of α-amylase in barley aleurone layers and this phenomenon appears to be due to an increase in α-amylase RNA. High amylase activity was reported in standard height genotypes than in short statured ones and a direct relationship between amylase activity and plant height was reported only in standard height genotypes in wheat[3]. In short statured genotype group, on the other hand, no relationship was discernible between plant height and amylase activity in endosperm during germination. Here we report some observations made on amylase activity of endosperms during germination in pearl millet seedlings in relation to dwarfism.

Seeds of seven cultivars of bajra [*Pennisetum americanum* (L.) Leeke] with a wide range of plant height at maturity (113–253 cm) were obtained from Millet Specialist, Gujarat Agricultural University, Jamnagar, India. Seeds of uniform size were surface-sterilized with 0.1% $HgCl_2$ for 10 min. They were then thoroughly washed with tapwater followed by several washings with distilled water. The washed seeds were placed over a moistened filter paper in petri dishes and incubated in the dark room (25±2°C). After 24 h, the uniformly germinated seeds were transferred to 9 cm petri dishes containing 5 ml of nutrient solution[4] and were transferred to room light. Seedlings were harvested at 24, 48, 72 and 96 h and dissected-out endosperms were homogenized in 3 ml of chilled sodium phosphate buffer (10 mM, pH 6.4). The homogenate was centrifuged at 3000 *g* for 15 min in cold. The supernatant served as the source for cytoplasmic fraction and the residue was used for the extraction of the wall bound fraction of amylase. The cytoplasmic fraction was purified by using chilled aqueous acetone (1 : 2). The precipitated proteins were resuspended in 10 ml extraction buffer. The residue, used for the extraction of wall-bound amylase, was washed several times with extraction buffer till there was no cytoplasmic amylase activity. Sodium chloride solution (1 N, 5 ml) was added to the residue and kept at room temperature for 45 min with frequent stirring to release wall bound enzyme. The extraction was repeated twice and all the supernatants pooled together, formed the source for wall bound amylase enzyme.

Total amylase activity was assayed following the procedure described by Jones *et al*[5]. The values of amylase activity were calculated and expressed as μg starch hydrolysed min^{-1} (mg protein)$^{-1}$. The protein content in cytoplasmic and wall bound enzyme fractions were determined using Folin reagent[6].

Cytoplasmic amylase activity in endosperm increased (200–500%) with seedling growth up to 72 h there after it showed a decrease. Amylase activity at 24, 48 and 72 h showed a direct relationship with mature plant height (figure 1a). In all the cultivars the maximum amylase activity was recorded at 72 h of growth. The decrease after 72 h may be due to the depletion of reserves in the endosperms. Wall bound amylase in endosperms did not reveal any clear trend during seedling growth. This enzyme activity showed a direct relationship with mature plant height only at 24 and 96 h (figure 1b).

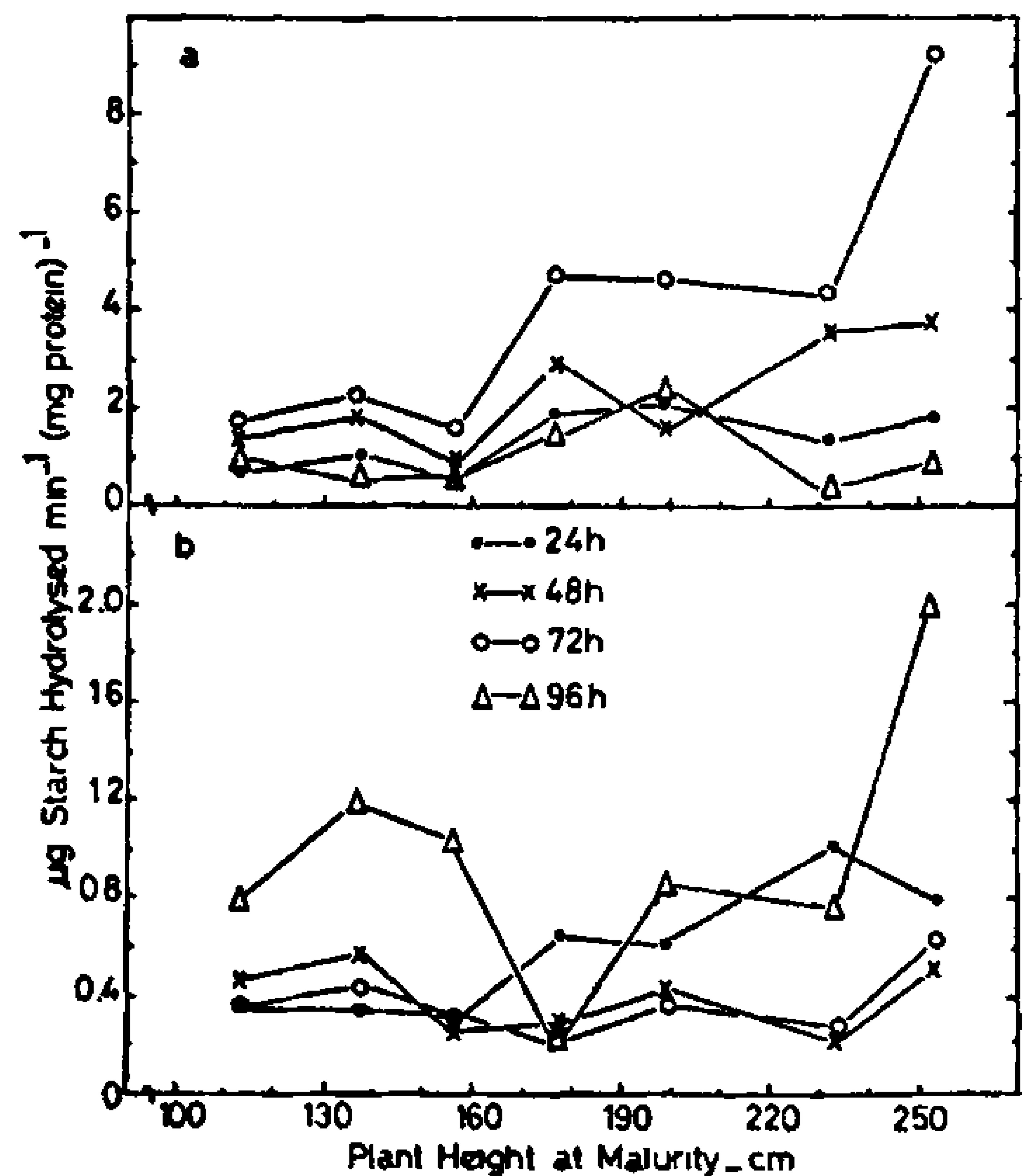

Figure 1. Changes of cytoplasmic (a) and wall bound (b) endosperm total amylase activity in seven cultivars of bajra varying in mature plant height during early seedling growth.

Role of GA in mobilization of reserve materials from endosperm of growing axis is well established[7,8]. That the development of amylase activity in incubated deembryonated *Zea mays* kernels occurred independently of the presence of embryo and did not require an exogenous source of GA has also been noted. Hence it was concluded that the inhibition of hydrolysis of the reserves following germination might be due to the release of hydrolyses preformed in the endosperm during maturation or that the endosperm contained sufficient endogenous GA to stimulate maximum hydrolyses production. The effect of added GA_3 on maize endosperm varied in different hybrids and inbred lines, because some already had a higher endogenous GA level while, others, e.g. dwarf maize are naturally deficient in GA[9]. The latter showed 3–5 fold increase in hydrolase production by GA application. In the present study cytoplasmic amylase activity in endosperms recorded a direct relationship with mature plant height indicating clearly that GA biosynthesis or its levels may be higher in tall cultivars than in dwarf ones; hence increased amylase activity in endosperms of tall cultivars than dwarf ones.

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INCREASED FREQUENCY OF NORMAL PLANT REGENERATION FROM CROWN GALL CALLUS OF *SESBANIA ROSTRATA*

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REGENERATION of plants from callus cultures of leguminous species has been found to be difficult. A few exceptions are the reports on pea[1], soybean[2] *Vigna aconitifolia*[3] and clovers[4]. Currently there is wide interest in *Sesbania rostrata* an annual legume which in addition to the root nodules, produces nitrogen fixing nodules on the stem and the branches[5]. Tissue culture and plant regeneration experiments with *S. rostrata* have been reported recently[6]. Though callus induction from a variety of explants was easy, plant regeneration from callus was observed at a low frequency. We also observed easy induction of callus in leaf and stem explants of this species. In the course of these experiments, regeneration of normal, non-transformed, nopaline negative shoots from axenic *Agrobacterium tumefaciens* induced tumour tissue was observed in over 50% of the cultures. These results are reported in this paper.

Seeds of *S. rostrata* were treated with concentrated H_2SO_4 for 30 min and then surface-sterilized using 0.1% $HgCl_2$. After repeated washing with sterile water these were cultured on Murashige and Skoog[7] medium. One-month-old seedlings were the source of leaf and stem explants. MS basal medium with 6-benzyl-adenine (BA) and α-naphthalene-acetic acid (NAA) were used in various experiments. Medium was adjusted to pH 5.8 before solidifying with 0.8% Difco agar.

A. tumefaciens strain A208 containing pTiT37 was used for inducing crown galls on the plants grown in the green house. The in planta tumours were cultured *in vitro* on MS medium supplemented with 200 mg/l Claforan. The regenerated shoots were induced to root on MS + 0.1 mg/l NAA. All cultures were kept at a light intensity of 8.2 watts/m^2 at $25 \pm 2°C$.

The rooted plants were transferred to autoclaved soil and later to pots in the field net house. Tumour tissue and the shoots arising from axenic cultures were checked for the presence of nopaline as described earlier[8].

The *Agrobacterium* inoculated sites on stem produced tumours within ten days which gradually enlarged (figure 1). The tumour tissue could be cultured on MS medium alone without the addition of auxin or cytokinin and showed profuse growth (figure 2).

The response of leaf, stem and tumour explants cultured on MS and MS supplemented with BA and NAA is shown in table 1. Leaflets and stem segments showed scanty callus growth on MS medium. Profuse callus growth in these explants was observed in MS + BA + NAA medium.

Table 1 *Response of leaf, stem and tumour explants of Sesbania rostrata*

Medium/explant	Stem	Leaf	Tumour
MS	C^+	C^+	C^{++}
MS + 1.5 mg/l BA +	C^{++}	C^{++}	C^{++}
0.1 mg/l NAA	ShR 1/48	ShR 0/48	ShR 26/48

Note: Number of culturers 48; Culture period 40 days; C^+ scanty callus; C^{++} profuse callus; ShR number of cultures showing shoot regeneration.