or (c) slightly longer (middle stigma only) than lemma as in well developed ones. Only in such cases at least a portion of stigma comes out on opening of floret.

The stigma is usually two in number, the third one is not well developed. The entire stigmatic portion is only 3–4 mm long. Tip of middle stigma of 1.5–2 mm struggles, along with stamens, to come out when lemma and palea widen. Rest of stigma remains inside the closed floret. The receptive stigmatic portion exposed to receive pollen is thus very limited.

The bristle-like hairs of palea (Figure 1 *a*) are longer than the stigmatic hairs and in an opened floret the exposed but small stigmatic portion is often covered (?) by the hairs on the two keels of palea. Possibly this can act as a barrier preventing stigma from receiving the pollen grains.

Moreover, the reported[10] higher percentage of sterility (70–92%) of pollen may also contribute to the sterility in *B. vulgaris*.

The available evidence points to the imminent danger of extinction of this mysterious species due to (i) the death of clumps after flowering, (ii) the lack of fruit set, (iii) the inherent 'unhealthy' nature of stigma to receive pollen, (iv) the possible role of bristle-like hairs as barriers preventing pollination, and (v) the high pollen sterility.

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Restriction fragment length polymorphisms of the rRNA genes in some pulses

Recognition and exploitation of variations among genetically divergent groups of germplasm are fundamental in breeding and genetic engineering programmes. Restriction fragment length polymorphisms (RFLPs), random amplified polymorphic DNAs (RAPDs), DNA finger printing, inter simple sequence repeat amplification (ISSR) and amplified fragment length polymorphisms (AFLPs) are powerful tools for studies of plant genetics, evolution, germplasm diagnosis and crop improvement[1–4]. These techniques allow a direct analysis of the plant genome at the DNA level. RFLP analyses have been used as molecular markers to construct linkage maps of crop plants, to mark quantitative trait loci and to complement phylogenetic relationships in several plant taxa[5–7]. rRNA genes, although not with the same impact as chloroplast DNA, have proven to be of tremendous utility in phylogenetic reconstruction[8]. They also provide valuable genetic markers for the analysis of genomic relationships among cultivated species and their wild relatives[9]. The tandem arrays of rDNA repeat units, generally located in the nucleolar organizing regions (NORs) of chromosomes, combine highly conserved gene regions, encoding rRNA, with more variable intergenic spacer regions (IGS). IGS regions, which separate the adjacent transcription units, have been found highly variable in sequence, length and copy number of subrepeats in several plant genera[10]. Because rRNA gene sequences are subjected to relatively rapid rates of concerted evolution[11], they produce DNA fragmentation patterns that are highly homogeneous within individuals and among closely related populations or species, yet exhibit characteristic heterogeneity between groups. In comparison, single copy gene markers[12] often tend to exhibit as much within-group as between-group variation in plant species. rRNA polymorphisms can, therefore, constitute useful genetic markers[9,13]. To achieve a better understanding and to provide molecular evidence for the systematic relationship between and within some populations of *Lablab*, *Dolichos* and *Vigna* species we investigated the polymorphism of the rRNA genes.

Five populations of *Lablab purpureus*,

one population each of *Dolichos trilobus*, *Vigna bournii*, *V. grahmiana*, *V. unguiculata* and *V. wightii* were used for the study. These natural populations were collected from Pulney Hills of Western Ghats, Tamil Nadu, India. The voucher specimens are deposited at Rapinat Herbarium, Tiruchirapalli (RHT), India.

After sacrification seeds were soaked in water overnight (10–16 h), rinsed thoroughly and grown on wet vermiculite under a light bench (10,000 lux, day/night cycling: 16 h/8 h). After three days the seedlings were planted in pots containing garden soil and grown. After 30 days, the primary leaves were harvested, frozen with liquid nitrogen and stored at –70°C until DNA extraction.

Total plant DNA was isolated and purified using a combination of the maize DNA miniprep method[14] and the cetyltrimethyl ammonium bromide (CTAB) method[15], as generally suggested[16]. The yield of DNA was 30–100 μg g^{-1} tissue; the UV absorbance ratios at 260 nm/280 nm and 260 nm/230 nm, were at least 1.9 and 2.1 respectively.

All DNA samples (5 μg each) were digested with *Eco*R1 and *Bam*H1, (5 units per μg DNA) in buffers recommended by the manufacturer (Boehringer Mannheim) at 37°C overnight. Electrophoresis was performed following standard procedures[17]. The molecular size marker Raoul-1 (Appligene, France) was used. Capillary blotting of the gels to nylon membranes (Hybond N, Amersham) was done according to the instructions of the manufacturer. A 3.5 kb *Bam*H1 fragment of the genomic clone PTA 250 (ref. 18) was used in this study. It was labelled with digoxigenin (Boehringer Mannheim). Prehybridization, hybridization and washing of the membranes were done following usual methods[17]. Detection was done nonradioactively with chemiluminescence. For autoradiography, the X-ray film (X-OMAT AR, KODAK) was exposed to the membranes between two intensifying screens (Lighting-Plus, Du Pont) at –70°C for 5 h.

Inter-generic, inter-specific and intra-specific polymorphism could be detected with the two endonucleases used in this study (Figure 1 *a* and *b*). Polymorphisms between species are higher than polymorphisms within species or between genera. The results show that only a few polymorphisms could be detected within the coding regions of the rRNA genes of *Lablab purpureus* whereas there were greater number of polymorphisms between species and between genera. These data correspond with results found in other plants[19].

The hybridization patterns of five populations of *Lablab purpureus* showed great similarity. The hybridization patterns of four different species of *Vigna* were very dissimilar. The hybridization patterns of the three different genera, namely *Lablab*, *Dolichos* and *Vigna* were also quite dissimilar. Our RFLP analysis supports the taxonomic classifications[20,21]. The appearance of clear bands suggests a limited number of loci, each containing a small or large number of repeats.

The present study on rRNA genes reflects the relatedness among genomes of the *Vigna* species with reference to the restriction sites.

Our results indicate that RFLPs of the rRNA genes are helpful in detecting inter-generic, inter-specific and intra-specific variations and thus contribute to an understanding of taxonomic relationships.

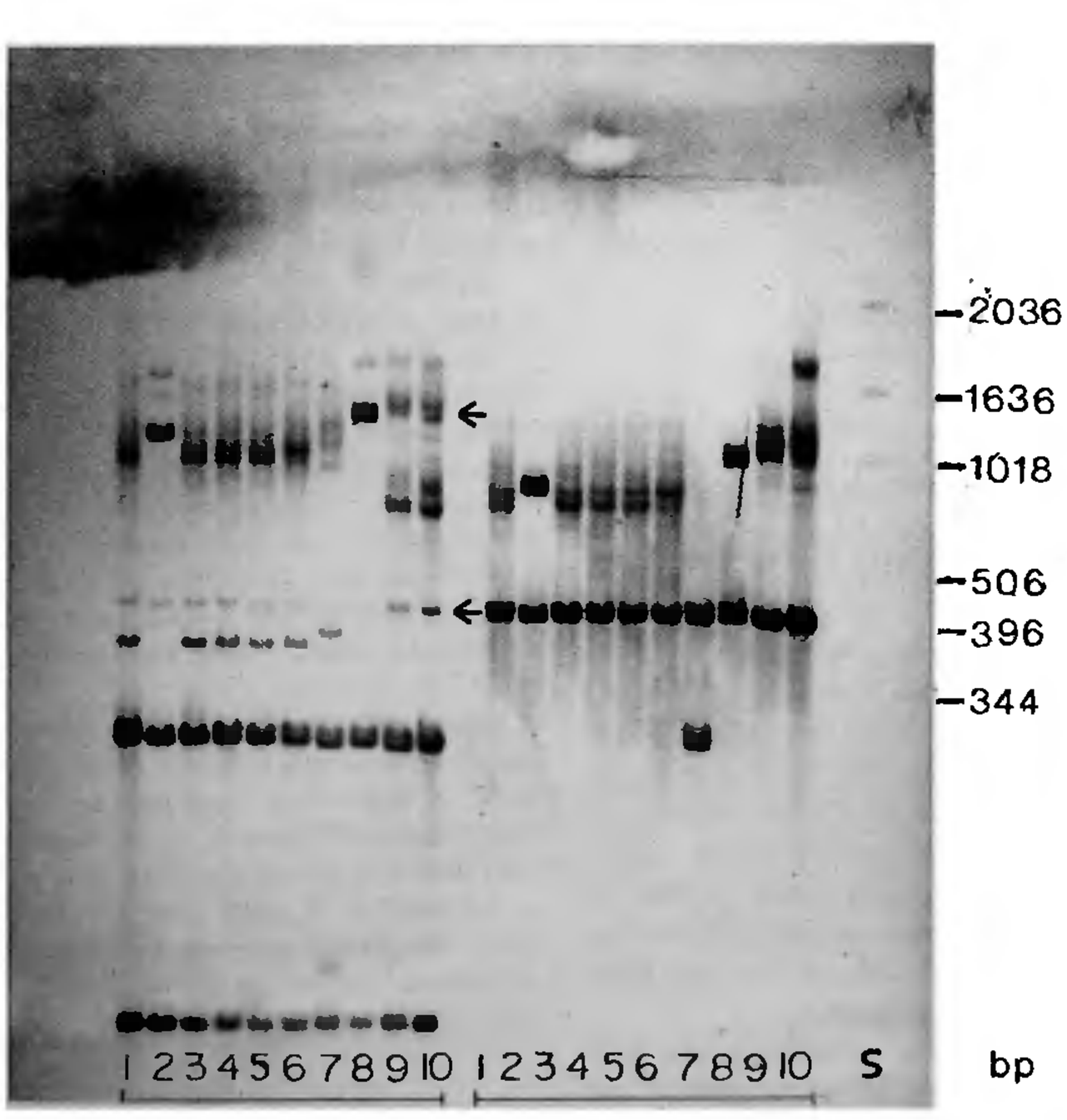

Figure 1. 1. *Lablab purpureus* population 1; 2. *Dolichos trilobus*; 3. *Lablab purpureus* population 2; 4. *Lablab purpureus* population 3; 5. *Lablab purpureus* population 4; 6. *Lablab purpureus* population 5; 7. *Vigna bournii*; 8. *Vigna grahmiana*; 9. *Vigna unguiculata*; 10. *Vigna wightii*; S = Standard Marker. *a* = *Eco*R1 digested RFLP patterns. *b* = *Bam*H1 digested RFLP patterns. Arrows = intergenic spacer region.

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Height vs water depth for small sand ripples – An aid to palaeohydraulics

Water flowing over a plane non-cohesive sediment surface throws it into a series of rhythmic bedforms which are designated as ripples (height < 0.075 m) and dunes (≥ 0.075 m) in lower flow regime. The nature and magnitude of these bedforms of lower flow regime are dependent on parameters like flow velocity, water depth and sediment grain size. The relationship between dune height and water depth has been worked out by Allen[1], $H = 0.086 \cdot d^{1.19}$ (H being the dune height and d, the corresponding water depth). Using some well-known hydraulic relationships in proper sequence, attempts are made to determine the flow velocities and mean annual discharge for ancient streams from dune heights preserved in rock sequences[2–4] (see ref. 5, pp. 279–280 for details).

The dimensions of the dunes used by Allen for establishing the above-mentioned relationship are large (H varying between 0.10 m and 10 m), but the ripples encountered in ancient sediments (rock record) are often of much smaller dimension. A series of experiments were conducted in a 'close circuit' hydraulic channel using sand grains of two different size ranges (Table 1) to determine the relationship between small ripple height and water depth. The heights and wavelengths of the ripples generated over these sand beds at a fixed flow velocity (~ 0.30 m/s) but varying water depths (0.088–0.185 m above the sand bed) ranged between 0.009–0.0165 m and 0.17–0.32 m respectively. The relationship between water depth and ripple height obtained from these experiments is $H = 0.065 \cdot d^{0.82}$, d being the effective water depth above the ripple crest (Figure 1). This relationship, somewhat different from that obtained by Allen, is expected to provide a realistic clue to palaeohydraulic parameters when small ripples are involved. Obviously, this relationship is valid for small water depths only. Although Allen used water depths as large as 10 m, in great water depths (as in oceans) ripple height may be independent of water depth.

Table 1. Grain-size of the sands used in the experiments

Sand no.	Mean grain-size (mm)	Standard deviation (mm)
IIT-1.1 (*)	0.42	0.287
IIT-4.1 (+)	0.175	0.052

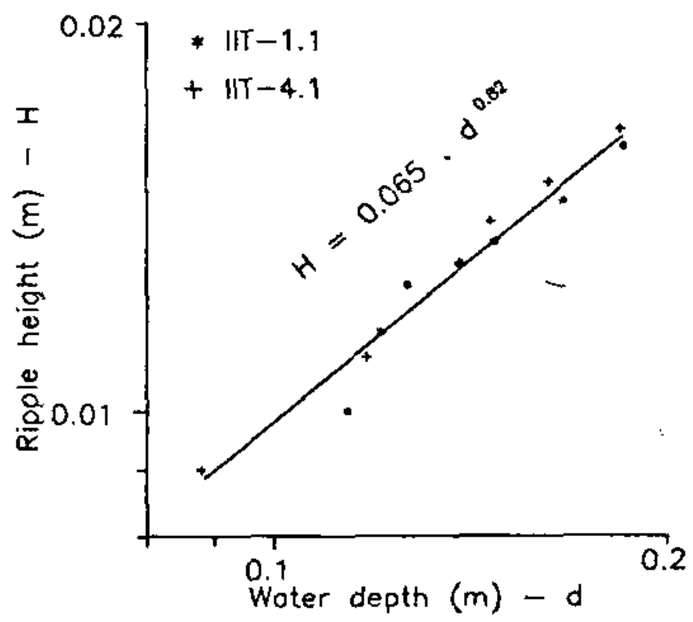

Figure 1. Water depth vs ripple height for small ripples.

The dimensions of subaqueous bedforms are known to be dependent not only on water depth, but also on flow velocity and grain size. The ranges of grain size and flow velocity for the data compiled by Allen are not explicitly stated in his publication. In the present case the grain sizes of both the sand samples used for the experiments as also the flow velocity lie within the lower flow regime ('ripple field' of Southard and Boguchwal[6]), thereby eliminating the possibility of distortions arising out of grain size or velocity variation.

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