terms of its available nitrogen, phosphorus and potassium content (Table 3). Results are in tune with Shyam¹⁴.

Results suggest that besides medicinal use, bloom of Ak can be successfully used for production of biogas with higher fertilizer value.

- Sastry, C. S. T. and Kavathekar, K. Y., Plants for Reclamation of Wasteland, Publication and Information Directorate, CSIR, New Delhi, 1990, pp. 175–179.
- Caius, J. F., The Medicinal and Poisonous Plants of India, Scientific Publ., Jodhpur, 1986.
- Das, B. B., Rasraj Mahodadhi, Khemraj Shri Krishnadas Prakashan, Bombay, 1996.
- 4. Oudhia, P. and Tripathi, R. S., World Weeds, 1999, 4, 109-119.
- Oudhia, P., Kolhe, S. S. and. Tripathi, R. S., Legume Res., 1997, 20, 133–136.
- 6. Oudhia, P., Kolhe, S. S. and Tripathi, R. S., Abstract. III International Congress

- on Allelopathy in Ecological Agriculture and Forestry, UAS, Dharwad, 18–21 August 1998, p. 151.
- Oudhia, P., Kolhe, S. S. and. Tripathi, R. S., First International Agronomy Congress on Agronomy, Environment and Food Security for 21st Century, Vigyan Bhawan, New Delhi, 23–27 November 1998, p. 27.
- Mital, K. M., Biogas Systems: Principles and Applications, New Age International (P) Ltd, New Delhi, 1996.
- Khandelwal, K. C. and Mahdi, S. S., Biogas Technology: A Practical Handbook, Tata McGraw-Hill Publishing, New Delhi, 1986.
- Somayaji, D. and Khanna, S., World J. Microbiol. Biotechnol. (Hist. Arch.), 1994, 10, 521-523.
- Kaparaju, P. and Rintala, J., Resour. Conserv. Recycling, 2005, 43, 175– 188.
- Kivaisi, A. K. and Rubindamayugi, M. S.
 T., Renewable Energy, 1996, 9, 917–921.

- Callaghan, F. J., Wase, D. A. J., Thayanithy, K. and Forster, C. F., *Biomass Bioenergy*, 2002, 27, 71–77.
- Shyam, M., Energy Sustain. Dev., 2002,
 37-42.

Received 6 June 2006; revised accepted 5 October 2006

PRATEEK SHILPKAR^{1,*}
MAYUR SHAH¹
D. R. CHAUDHARY²

¹Biogas Research and Extension Centre, Gujarat Vidyapith, Sadra,

Gandhinagar 382 320, India
²Department of Phytosalinity,
Central Salt and Marine Chemicals
Research Institute,

Bhavnagar 364 001, India *For correspondence. e-mail: pshilpkar@yahoo.com

Studies on stem structure of X Laburnocytisus adamii (Poit) Scheid

X Laburnocytisus adamii (Poit.) Scheid. is a graft hybrid of Laburnum anagyroides Medik. and Cytisus purpureus Scop., both belonging to the family Fabaceae. Adam, a nurseryman from Vitry near Paris, inserted a shield from the bark of a lowspreading, purple-flowered shrub C. purpureus (purple broom) into a stock of yellow-flowered L. anagyroides (golden chain tree). The bud lay dormant for a year and then grew upright and vigorous with larger leaves than is usual for broom. This graft hybrid continued to grow, the main limb producing foliage and long drooping chains of yellow flowers along with clumps of purple flowers on twiggy branches and pinkish flowers on other longer branches.

The yellow and purple flowers produced by *X L. adamii* were similar to those of the parent plants, while the pink flowers had characteristics intermediate between the two originally grafted plants. The pink flowers were associated with somatic fusion of cells of the two graft partners, while the revertant sectors (i.e. branches that formed flowers identical to either of the originally grafted plants) were the result of segregation of the characters back to uniformity. Such a

graft hybrid, also called 'chimera', is a bigeneric hybrid, which has originated artificially as a result of grafting of two different genera^{1,2}.

Here we compare the stem structure of shoots associated with the three different types of flowers found in *X L. adamii*, and see whether the *Laburnum*-type and *Cytisus*-type differed in stem structure from *L. vossii* and *C. purpureus* respectively.

Shoots were selected from the *X L. adamii* tree growing in the Treborth Botanic Garden, UK, having leaf and flower types characteristic of *Laburnum*, *Cytisus* and *X Laburnocytisus*. Shoots were also selected from plants of *C. purpureus* and *L. vossii* (a close relative of *L. anagyroides*). Samples from these shoots were pickled and preserved in formalin–propionic–alcohol (FPA). These samples were washed in tap water before cutting sections, and portions remaining after sectioning were preserved in 70% alcohol.

Sections were cut by hand with a singleedged razor blade, in transverse, tangential longitudinal and radial longitudinal directions. These were mounted in a dilute solution of iodine in potassium iodide (which stains starch grain black and acts as a general differential stain for cellulose and lignin). These temporary mounts were examined and photographed immediately with a transmitted light microscope. Polarized light was also used to show up any crystals present.

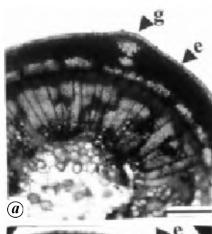
The three types of shoot from the *X L.* adamii tree are described here as *Laburnum*-type, *Cytisus*-type and *Laburnocytisus*-type.

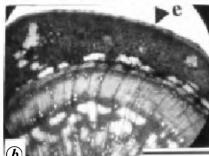
Laburnum-type shoots were similar to those of L. vossii in having a rounded first-year stem, with slight ridges on each side of the petiole base, but these faded out a short distance below the leaf axil. The young first-year stems of both Laburnum-type and L. vossii were covered in white hairs closely adpressed to the stem, but these soon disappeared leaving a green stem. Later, in the season the epidermis turned pale brown, then broke up to show a smooth and shiny green periderm below.

In contrast, *C. purpureus* and the *Cytisus*-type stems both had persistent longitudinal ridges on either side of each petiole, continuing up to the petiole above: these ridges became less obvious in third-year stems. The bark of older *C. purpureus* and *Cytisus*-type stems was rougher than in the *L. vossii and Laburnum*-type, with

elongated vertical ridges: that of the *La-burnocytisus*-type appeared intermediate.

The differences in external appearance between the stem types were associated with differences in the rate of change from the original epidermis to production of a periderm. Successive stages in the change from epidermis to periderm are shown in Figures 1 a–c, 2 a, b and 3 a, b. The multilayered periderm was produced within the cortex, and the epidermis and





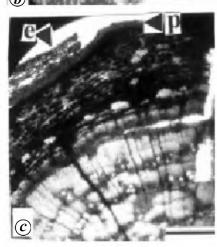


Figure 1. Transverse sections of *Cytisus* purpureus stems. Scale bar 1 mm. a, One-yrold stem. g, Group of gelatinous fibres beneath ridge on bark and e, Epidermis. b, Two-yr-old stem. e, Epidermis. c, Three-yr-old stem. e, Epidermis and outer cortex beginning to strip-off and p, Periderm exposed where outer tissue stripped-off.

outer cortex layers subsequently peeledoff. This happened much earlier in *La-burnum*-type and *L. vossii* stems (in the first year) than in *Cytisus*-type and *C. purpureus* stems (in the second or third year).

In surface view, the epidermis included stomata in all shoot types, plus hairs in *Laburnum*-type and *L. vossii* (or hair base where the hairs had broken-off), but not in *Cytisus*-type or *C. purpureus*.

In surface view, the periderm had thick-walled polygonal cells in *Laburnum*-type, *Laburnocytisus*-type and *L. vossii* (Figure 4 a). In sections, the periderm cells appeared to be thinner-walled in *Cytisus*-type and *C. purpureus* (Figure 4 b) than in *L. anagyroides, Laburnocytisus*-type and *L. vossii* (Figure 5 a-c). The periderm cells were arranged in neat rows (radially and longitudinally), with the outer periclinal walls particularly thickened (Figure 5 a and c).

Beneath each bark ridge there was a group of gelatinous fibres (Figures $1\,a$, $2\,a$), which appeared to originate as cap cells to vascular bundles from the leaf petiole. The gelatinous bark fibres were in blocks in the outer bark of all stem types: these blocks appeared to be more

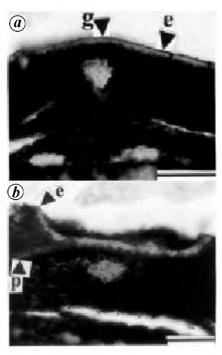
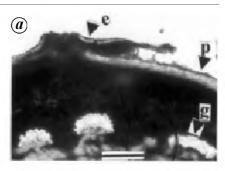


Figure 2. Transverse section of *Cytisus*-type shoots from *X Laburnocytisus adamii. a*, Scale bar 0.3 mm. g, Group of gelatinous fibres beneath ridge on bark and e, Epidermis. b, Scale bar 0.2 mm. e, Epidermis beginning to break up and p, Periderm developing in cortex



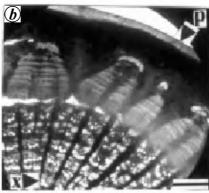
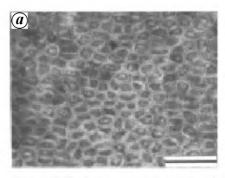


Figure 3. Transverse section of *Laburno-cytisus*-type shoots from *X L. adamii*. Scale bar 0.3 mm. e, Epidermis beginning to break up and g, Cap of gelatinous bark fibres. *b*, Scale bar 0.3 mm. p, Periderm and x, Xylem with clusters of vessels, group of gelatinous fibres and multiseriate rays.



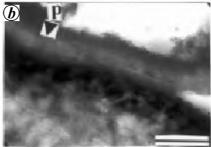
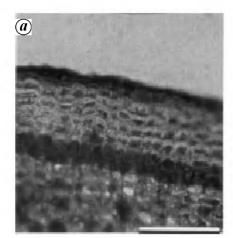


Figure 4. a, Laburnocytisus-type shoots from X L. adamii showing surface view of periderm cells with thick walls. Scale bar 0.1 mm. b, Transverse section of C. purpureus stem. Scale bar 0.1 mm. p, Periderm with thinner cell walls than in X L. adamii or L. vossii.

conspicuously crescent-shaped in Laburnocytisus-type and L. vossii (Figure 3 a and b) than in Cytisus-type or C. purpureus (Figure 2 a, b). Gelatinous fibres have been reported in other species of Leguminosae like Black Locust, Robinia pseudoacacia as a result of tension wood³. The presence of gelatinous fibres in reaction tissue of family Leguminosae has

vossii (Figure 6 a and b). In Laburnocytisus-type, Cytisus-type and C. purpureus, the stone cells appeared to be less frequent with scattered individual cells or small groups in the cortex (Figure 6c).

also been reported⁴. Large groups of stone cells were found in the cortex of Laburnum-type and L.





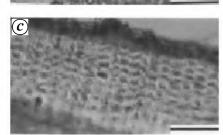
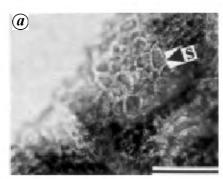


Figure 5. a, Radial longitudinal section of Laburnocytisus-type shoots from X L. adamii showing multilayered periderm with cell thickened on their outer periclinal walls. Scale bar 0.1 mm. b, Transverse section of L. vossii stem showing multilayered periderm in regular rows. Scale bar 0.1 mm. c, Transverse section of Laburnum-type shoots from X L. adamii showing multilayered periderm. Scale bar 0.1 mm.

Rays inflated in the bark in all stem types (Figures 1c and 3b), so that the phloem tended to be in triangular blocks.

Little difference was detected between stem types in overall wood structure. All types were ring porous (Figure 7a). These results are in conformity with the findings of Werner et al.⁵ in L. anagyroides. Latewood vessels clustered in groups, tending to dendritic or tangential in arrangement (Figure 7 a). Scattered paren-



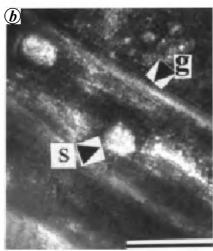


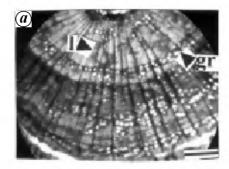


Figure 6. a, Tangential longitudinal section of L. vossii stem. Scale bar 0.1 mm. s, Group of thick-walled stone cells in bark. b, Tangential longitudinal section of Laburnum-type shoots from X L. adamii. Scale bar 0.3 mm. g, Gelatinous bark fibres and s, Group of thickwalled stone cells, c. Radial longitudinal section of C. purpureus stem. Scale bar 0.1 mm. s. Individual thick-walled stone cells.

chyma was present around the vessel groups, and storied tissues were also present. Axial parenchyma has been reported in Laburnum and Cytisus by Cutler et $al.^6$.

In all stem types, vessels appeared similar in structure: round in transverse section, thick-walled and well-pitted. In all types the bordered pits were alternate, and round, oval or slightly elongated (Figure 7b). Spiral thickening was present in some vessels (Figure 7 b).

Rays were possibly wider in Laburnum than in Cytisus-type (though this may be age-related). In all stem types ray cells were rounded in tangential longitudinal section (Figure 8 a and b). In radial longitudinal section of all stem types, the ray



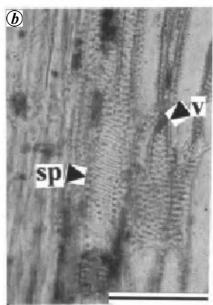
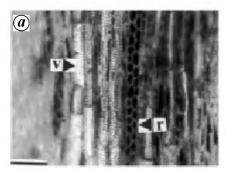


Figure 7. a, Transverse section of Cytisustype shoots from X L. adamii. Scale bar 0.3 mm. gr, Growth ring boundary showing ring of earlywood vessels and l, Latewood vessels in dendritic arrangement. b, Radial longitudinal section of Laburnum-type shoots from X L. adamii. Scale bar 0.05 mm. sp, Spiral thickening in vessel, and v, Alternate pitting in vessel.

cells had thick, well-pitted walls (Figure 9 a and c). Rays were heterogenous, but there appeared to be a higher proportion of procumbent cells in *Laburnum*-type and *L. vossii* (Figure 9 a), compared to *Cytisus*-type, which had more upright cells (Figure 9 b and c), though this could also be related to the age or width of the rays.

Thus the main differences detected between the three different shoot types on *X L. adamii* were: (i) Presence of hairs on young shoots in *Laburnum* but not *Cytisus*-type and presence of bark ridges with longer persisting epidermis in *Cytisus*-than *Laburnum*-type. (ii) Crescent-shaped caps of gelatinous fibres in the cortex of *Laburnum*-type and *Laburnocytisus*-type; less obvious in *Cytisus*-type. (iii) Stone cells in groups in *Laburnum*-type; present as isolated cells in *Cytisus*-type and



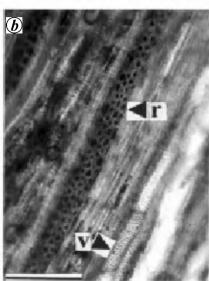
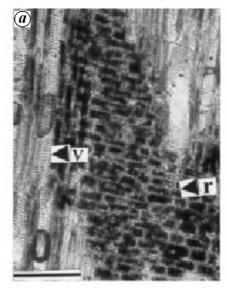


Figure 8. a, Tangential longitudinal section of C. purpureus stem. from X L. adamii. Scale bar 0.05 mm and r, Ray and v, Alternate pitting in vessel. b, Tangential longitudinal section of Laburnocytisus-type shoots from X L. adamii. Scale bar 0.05 mm. r, Multicelluar ray with cells rounded in TLS and v, Alternate pitting in vessel.

Laburnocytisus-type. (iv) Rays heterogenous in all specimens, but there appeared to be more procumbent than square or upright cells in Laburnum-type compared with more square and upright cells than procumbent in Cytisus-type. This needs conformation with further samples.

In all these features, the *Laburnum*-type stems appeared identical to those of *L. vossii*, and the *Cytisus*-type stems were



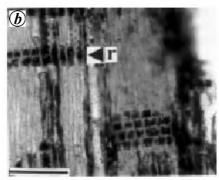




Figure 9. *a*, Radial longitudinal section of *Laburnum*-type shoots from *X L. adamii*. Scale bar 0.05 mm. r, Ray cells predominantly procumbent and v, Alternate pitting in vessel. *b*, *c*, Radial longitudinal section of *C. purpureus* stem. Scale bar 0.05 mm. (*b*) r, Ray cells either square or upright. (*c*) r, Ray cells with thick, well-pitted walls.

indistinguishable from those of *C. pur-pureus*. The *Laburnocytisus*-type stems appeared to be intermediate, veering towards the *Laburnum*-type in having crescent-shaped caps of gelatinous fibres, but towards the *Cytisus*-type in frequency of stone cells.

- Darwin, C., Variation of Animals and Plants under Domestication, Parts I, II, London, Murrey, 1868.
- Swingle, C. F., J. Hered., 1927, 18, 73– 94.
- Okuyama, T., Yamamoto, H., Yodhida, M., Hattori, Y. and Archer, R. R., Ann. Sci. For., 1994, 51, 291–300.
- 4. Hoster, H. R., BerlinDtsch. Bot. Ges., 1966, 79, 211-212.
- 5. Werner, S., Iris, H. K., Fritz, S. and Felix, K., http://www.woodanatomy.ch.22, 2004.
- Cutler, D. F., Rudall, P. J., Gasson, P. E. and Gale, R. M. O., Root Identification Manual of Trees and Shrubs. A Guide to the Anatomy of Roots of Trees and Shrubs Hardy in Britain and Northern Europe, Chapman and Hall, London, 1987, p. 244.

ACKNOWLEDGEMENTS. We thank Nigel Brown (curator of the Treborth Botanic Garden, UK) for help during this work. We also thank Marishal Thompson and Company Ltd. at the European Plant Science Laboratory, Parc Menai, Bangor, UK for use of their laboratory facilities for anatomical work. We are grateful to the Vice-Chancellor, Dr Y. S. Parmar University of Horticulture and Forestry, Solan, for providing leave to P.T. to conduct this research work during May—September 2005 in Treborth Botanical Garden.

Received 5 November 2005; revised accepted 11 October 2006

PRIYANKA THAKUR^{1,*}
KULWANT RAI SHARMA¹
MARY PAT DENNE²
AJAY THAKUR²

¹Dr Y.S. Parmar University of
Horticulture and Forestry,
Nauni, Solan 173 230, India
²School of Agricultural and
Forest Sciences,
University of Wales,
Bangor, United Kingdom
*For correspondence.
e-mail: priyanka.thakur@gmail.com