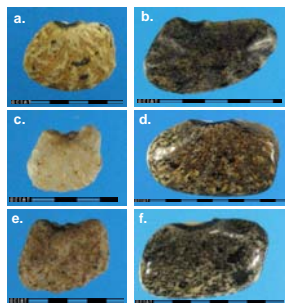


J.M. Salcedo<sup>1</sup>, N.P. Castañeda<sup>1</sup> & D.G. Debouck<sup>2</sup>

<sup>1</sup> Bioviversity International, Oficina para las Américas, c/o CIAT, COLOMBIA. [j.salcedo@cgiar.org](mailto:j.salcedo@cgiar.org)

<sup>2</sup> Centro Internacional de Agricultura Tropical (CIAT), AA 6713, Cali, COLOMBIA. [d.debouck@cgiar.org](mailto:d.debouck@cgiar.org)  
(work supported in part by BMZ of Germany)

Many wild *Phaseolus* species, belonging to sections as different as *Acutifolii*, *Falcati*, *Minklersia* and *Xanthotricha* (Freytag & Debouck 2002), have small seeds (100-seed weight: 0.5-3 g). This may be part of a *R* reproductive strategy, where photosynthesis carbohydrates are invested over weeks in larger numbers of smaller propagules instead of smaller numbers of larger seeds produced in a shorter period. This reproductive strategy might be appropriate in open, sunny, sometimes degraded (oak or pine) grasslands where many such species are distributed. Among the small-seeded species, there is a group with distinctive rugose testa, viz. the section *Rugosi* Freytag (Freytag & Debouck 2002), namely distributed in the Sonoran and Chihuahuan Deserts as currently understood (Dimmitt 2000, and Hernández & Gómez-Hinostrosa 2005, respectively). The *Rugosi* currently includes: *P. filiformis* Benth., *P. angustissimus* A. Gray, and *P. carteri* Freytag & Debouck. They seem also to form a distinct phylogenetic entity, remotely linked to the group of *P. vulgaris* (= the Phaseoli), on the basis of biochemical (Jaaska 1996), molecular (Skroch et al. 1993; Delgado-Salinas et al. 2006) and hybridological (Belivannis & Doré 1986) evidence. With help of scanning electron microscopy and geographic information systems, we were interested in testing further the validity of this section *Rugosi*. The *in situ* study of 38 herbaria holding specimens of the three above mentioned species gives us geographic and phenological information for 309 populations of *Rugosi* (Debouck 2009) (Figure 1).



A practical result of these testa surface projections is a net surface increase at microlevel. Because we suspect this increase to be related with a water uptake mechanism during and for germination, we did a comparison of water capture by the testa by putting 20 seeds per species in water for one minute followed by a strong shaking (Table 1).

Fig. 2. Seed testa of a: *P. angustissimus*; b: *P. microcarpus*; c: *P. carteri*; d: *P. lunatus* (wild); e: *P. filiformis*; and f: *P. vulgaris* (wild) (scale bar in mm).

Species	Seed dry weight and S.D.	Water uptake and S.D.	Tukey Test
<i>angustissimus</i>	304 (± 53)	10.5 (±3)	A
<i>carteri</i>	81 (± 15)	7.85 (± 1)	BC
<i>filiformis</i>	98 (± 17)	9.65 (± 2)	AB
<i>microcarpus</i>	152 (± 27)	4.50 (± 1)	D
<i>lunatus</i>	1131 (± 152)	7.20 (± 2)	C
<i>vulgaris</i>	380 (± 55)	4.60 (± 2)	D

Table 1. Means of seed dry weights and of water uptakes with respective standard deviations, and Tukey test (data in g E - 04).

An analysis of variance shows that there are significant differences ( $F = 30.6$ ,  $p < 0.001$ ), at a level of confidence of 95% for the water uptake by the seed testa. A relatively high coefficient of variation was observed for the seed dry and wet weights ( $CV = 19.83$  and  $27.36$ , respectively), because of the variation from one seed to another within each sample, but of the same order of magnitude, inferring thus a lack of bias towards anyone species. The Tukey test shows a significant effect of the rugose testa in the uptake of superficial water, clearly in favour of the *Rugosi*, as compared to the other species herein tested.

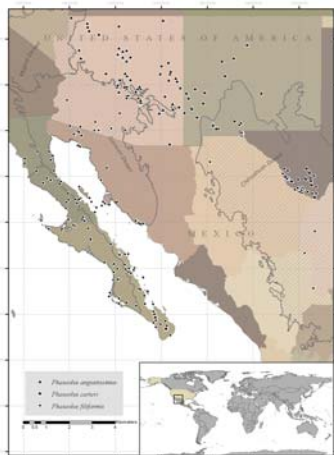


Fig. 1. Distribution map of species of section *Rugosi*.

## RESULTS AND DISCUSSION

Seed testa of the three species of section *Rugosi* and another three for comparison can be seen in Fig. 2a-f. As compared to the testa of *P. microcarpus* (Fig. 2b) and those of *P. lunatus*, which displays some veins irradiating from the hilum at the macrolevel (Fig. 2d), the testa of the *Rugosi* are indeed rugose ("tuberculate", according to Lackey 1983) at x 500 magnification (Fig. 2a, c, e). In contrast, the seed testa of *P. vulgaris* are completely smooth (Fig. 2f). But in addition to an undulate rough surface the *Rugosi* display spherical structures at x 2,500 magnification, usually mounted on small conical elevations, at a variable density, lower in *P. angustissimus* (Fig. 3a-b) and *P. carteri* (Fig. 3c-d) as compared to *P. filiformis* (Fig. 3e-f).

Such structures are absent in *P. microcarpus* (Fig. 3g-h), *P. lunatus* (Fig. 3i-j), and *P. vulgaris* (Fig. 3k-l).

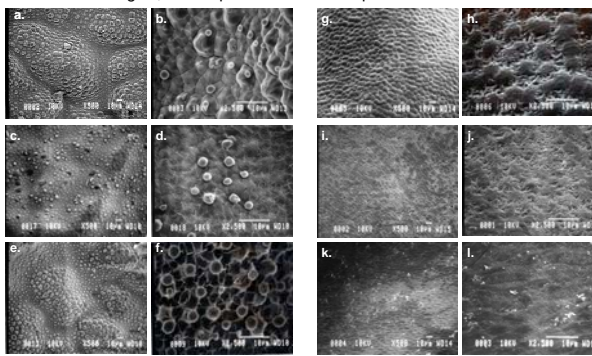


Fig. 3. SEM micrographs of seed testa of: a-b. *P. angustissimus*; c-d. *P. carteri*; e-f. *P. filiformis*; g-h. *P. microcarpus*; i-j. *P. lunatus* (wild); and k-l. *P. vulgaris* (wild).

The *Rugosi* species of *Phaseolus* are distributed in and around two deserts of North America: the Sonoran and the Chihuahuan Deserts (Dimmitt 2000) (Fig. 1). Because we suspect that the peculiar seed testa microtexture is an adaptation to dry environments, namely germination under low moisture availability in the soil, we checked the distribution of rainfall during the periods of favourable temperatures, at or close to the location of specific populations (Fig. 4a-e). Although data for *P. carteri* are few (Debouck 2009), this desert therophyte flowers in October, taking benefit from any of the wettest months (Jul-Sep; Fig. 4a). *P. filiformis* - with the highest density of microspheres - has a wide range of distribution from S Baja California up to W Texas, from sea level up to 2,000 m. As an opportunistic and short-cycle (it blooms at 20 days!) desert therophyte it takes benefit from the rains of Nov-Dec or Mar-Apr in lowland Baja California (Fig. 4b). As far as temperature is favourable, it also takes benefit from the scanty rains of Jul-Aug (45 mm / month) in highland Coahuila (Fig. 4c). *P. angustissimus* with a major root and shoot development takes benefit from the unimodal rainfall pattern in W Texas (Fig. 4d), overwintering as latent root. This root trait - combined with some tolerance to subzero temperature (Balasubramanian et al. 2004) - enables to take benefit from spring or fall rains in central W Arizona.

The *Rugosi* follow the general growth pattern of *Phaseolus* species with reproductive cycle under favourable temperatures and water availability. They do not enter thus in Mediterranean climates with the sole rainy season during the winter, and are naturally absent in California and Chile (Lackey 1983, and Freytag & Debouck 2002). The *Rugosi* have survived in the Sonoran and the Chihuahuan Deserts thanks to outstanding physiological attributes such as tolerance to low temperatures (Balasubramanian et al. 2004), and salinity (Bayuelo-Jiménez et al. 2002). But also as shown here thanks to a rugose testa very different from other small-seeded wild *Phaseolus* species: a corrugate testa with 'golf ball' microstructures that capture microdroplets of water, allowing a fast germination under water limited availability. Are the genes responsible for such microstructures 'silenced' in common bean?

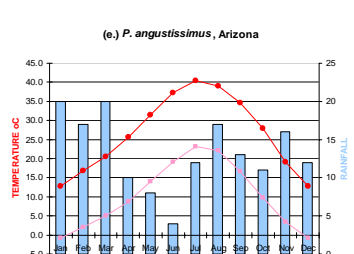
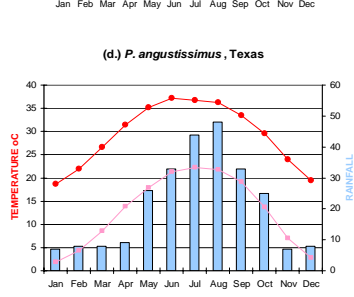
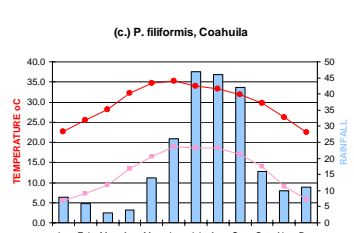
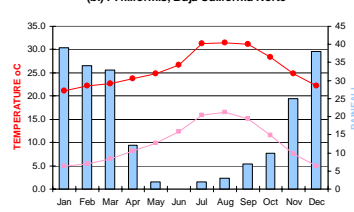
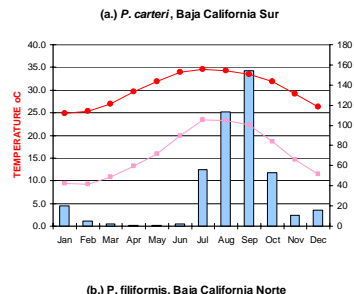


Fig. 4. Monthly distribution of rainfall (total mm/month) and temperature (minimum and maximum) for selected locations close to collection sites of: a. *P. carteri* (Brandege s.n.); b-c. *P. filiformis* (Villareal 4418 and Wiggins 4801, respectively); d-e. *P. angustissimus* (Barneby 18093 and Warnock 652, respectively).

## REFERENCES

Balasubramanian P., A. Vandenberg, P. Hued & L. Gusta. 2004. Resistance of *Phaseolus* species to ice crystallization at subzero temperature. *Physiol. Plant.* **120** (3): 451-457.  
 Bayuelo-Jiménez J.S., D.G. Debouck & J.P. Lynch. 2002. Salinity tolerance in *Phaseolus* species during early vegetative growth. *Crop Sci.* **42** (6): 2184-2192.  
 Belivannis, T. & C. Doré. 1986. Interspecific hybridization of *Phaseolus vulgaris* L. and *P. angustissimus* A. Gray using *in vitro* embryo culture. *Plant Cell Rep.* **5**: 329-331.  
 Debouck D.G. 2009. Cahiers de Phaséologie - section *Rugosi*. Consulted on line at: [publications.cgiar.org](http://publications.cgiar.org), on March 26, 2009.  
 Delgado-Salinas A., R. Bähler & M. Lavin. 2006. Phylogeny of the genus *Phaseolus* (Leguminosae): a recent diversification in an ancient landscape. *Syst. Bot.* **31** (4): 779-791.  
 Dimmitt M.A. 2000. Biomes and communities of the Sonoran desert region. In: "A natural history of the Sonoran desert", S.J. Phillips & P. Wentworth (eds.), Arizona-Sonora Desert Museum Press, Tucson, Arizona, USA, pp. 3-18.  
 Freytag G.F. & D.G. Debouck. 2002. Taxonomy, distribution, and ecology of the genus *Phaseolus* (Leguminosae-Papilionoideae) in North America, Mexico and Central America. *SIDA Bot. Misc.* **23**: 1-300.  
 Hernández H.M. & C. Gómez-Hinostrosa. 2005. Cactus diversity and endemism in the Chihuahuan desert region. In: "Biodiversity, ecosystems, and conservation in northern Mexico", J.L. Carrón, G. Caballero & R.S. Felger (eds.), Oxford University Press, New York, New York, USA, pp. 264-275.  
 Jaaska V. 1996. Isoenzyme diversity and phylogenetic affinities among the *Phaseolus* beans (Fabaceae). *Pl. Syst. Evol.* **200**: 233-252.  
 Lackey, J.A. 1983. A review of generic concepts and nomenclature of Phaseolinae (Fabaceae, Faboideae). *Iselya* **2**: 21-64.  
 Skroch P.W., R.C. Dobart, E.W. Triplett & J. Nianhui. 1993. Polymorphism of the leghemoglobin gene in *Phaseolus* demonstrated by PCR amplification. *Euphytica* **69** (3): 177-183.