A comparison of pod production and insect ratings of 12 elite *Prosopis alba* clones in a 5-year semi-arid Argentine field trial

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**A B S T R A C T**

Semi-arid adapted nitrogen fixing trees of the genus *Prosopis* have been extensively used in Argentina for soil improvement, luxury quality lumber production and sweet (35% sucrose) pods for humans and livestock. Due to the great demand of *Prosopis alba* for lumber, erect, tall, high pod producing trees of this species have been greatly overharvested, leading to erosion of the gene pool. A previous progeny trial with 57 half sib families identified 12 trees with promise for rapid growth, high production of pods and sweet pods. This trial, on a site with a salinity of 8.6 dS m$^{-1}$ EC and a pH of 7.7, examined clones of these 12 trees in a randomized complete block trial with 8 single tree replications for height, basal diameter, canopy height and diameter, production of pods, sensory characteristics of the pods, disease resistance and insect resistance. In the 5th year of production and 7th year from planting, three clones produced more than 50 kg pods per tree versus 32 kg for check. At this 10 m × 10 m spacing, this yield of 5000 kg/ha compares favorably to many other semi-arid crops, especially given the unfavorable salinity and pH. In contrast to genetic improvement in pod production, the clones had lower diameter, height and canopy growth than the check. The lower biomass production may be due to fibrous root system produced from cuttings, since some of the clones blew over in high winds but none of the checks produced from seed blew over. Companion seed orchards of salt tolerant clones may provide rootstock for these high pod production clones. Significant differences in insect and disease resistance of the clones were observed and full sib crosses were made to study the genetics of the resistance. All clones had good sensory properties for use in human food. This is the first replicated trial with multipurpose clones useful for lumber and human food. The annual diameter growth rates ranged from 2.8 to 4.1 cm year$^{-1}$ which was estimated to produce an internal return of approximately 20% from lumber alone.

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**1. Introduction**

Nitrogen fixing trees and shrubs of the genus *Prosopis* are an important feature of many of the worlds semi-arid ecosystems where they enrich the soil with organic N (Geesing et al., 2000), provide fuelwood to the local population, high sugar content pods for livestock and occasionally human use, and provide lumber for high quality furniture (Pasiecznik et al., 2001; Felker, 2009). Of all the *Prosopis* species, the most commercially valuable is *Prosopis alba* which has provided more than 100,000 tons of logs per year in the Province of the Chaco, Argentina for use in furniture, flooring, doors, and window coverings (M. Bejarano, pers comm, 2000). This extensive use for furniture is attributable to its radial, tangential and volumetric shrinkages of 1.8%, 2.9% and 4.8% respectively (Turc and Cutter, 1984) that are lower than all other hardwoods listed in the tropical timber compendium of Chudnoff (1984). The high sugar content pods of *P. alba*, ca 35% (Oduol et al., 1986) are highly palatable to livestock and humans and were a major source of food for indigenous peoples in Argentina (D’Antoni and Solbrig, 1977). As early as the 1940s, the eminent Argentine botanist Arturo Burkart (Burkart, 1976) decreed the selective overharvest of fastest growing, straightest trees and suggested the genetic improvement programs be initiated to select the best trees.

In the 1980s with funding from the Canadian International Development Research Council (IDRC) germplasm collections were made for four of the most economically important *Prosopis* species in Argentina i.e. *P. alba*, *P. chilensis*, *P. flexuosa* and *P. nigra* and half sib progeny trials established for *P. flexuosa* and *P. chilensis* near Mendoza, Argentina (Cony, 1996a,b) and *P. alba* near Santiago del Estero, Argentina (Felker et al., 2001).

In 1990 in Santiago del Estero, Argentina the *P. alba* progeny trial was established in a randomized complete block design with 57 families, 7 replications and 4 trees per replication. In 1997 and 1998 these trees were evaluated for height, basal diameter, and pod...
production. After 9 years, of the surviving 1289 trees in this trial, only 98 trees were more than 4 m in height and only 56 had produced more than 1.75 kg of pods year$^{-1}$. Of this group only 32 trees had more than 4 m in height and more than 1.75 kg pod year$^{-1}$.

As pods of some species, i.e. *P. juliflora* are so bitter/astringent they cannot be used for human food (Felker, 2009), pods of these 32 trees were ranked as being very bitter, bitter, sweet or very sweet and it was found that only 12 of these 32 trees had pods with a sweet or very sweet non-astringent taste. The 12 trees that possessed the best complement of economically useful characters i.e. that ranged in pod size from 6.55 to 14.4, 1.57 to 13.6 and 1.29 to 1.70 times the population mean of pod production (0.29 kg tree$^{-1}$), biomass (24.2 kg tree$^{-1}$) and height (3.1 m) respectively were propagated asexually (Felker et al., 2001).

In 2002, a randomized complete block trial with clones of these 12 trees was established in Argentina to compare their characteristics with a random seed propagated selection, to serve as a seed orchard and as breeding population. This paper compares the pod production of these clones to a random seed propagated check and reports initial ranking of insect and disease resistance.

### 2. Materials and methods

While cuttings can be rooted from fast growing, greenhouse grown *P. alba*, it is virtually impossible to root cuttings of mature field trees. Thus scions were taken from 12 mature elite trees in the progeny trial of 57 half sib families (Felker et al., 2001) and grafted by the techniques of Wojtusik and Felker (1993) onto greenhouse grown stock plants which were then used to produce rooted cuttings (Felker et al., 2005). A clonal evaluation/seed orchard was established in Fernandez, Argentina on grounds of the Universidad Catolica de Santiago del Estero (UCSE, Experiment Station (27°56′10.21″S, 63°52′37.73″W) on 21 January 2002. This is summer and the rainy season in this part of Argentina. The rainfall measured at the Research station in Fernandez was 771 mm in 2002, 410 in 2003, 709 in 2004, 402 in 2005, 750 in 2006, 674 in 2007 and 678 in 2008. The field was irrigated by flood irrigation of about 100 mm the first spring after planting on 23 September 2002.

Thereafter, every year (except for 2005 and 2006) in late winter (August) a similar single irrigation was made.

Based on data from a meteorological station at this site, generally every month from September (spring) through March (fall) has one day with a maximum temperature of 38 °C with yearly maximum temperatures of 43–46 °C occurring from December through February. The first significant rains normally occur in late October, and they end in mid April, with a peak in December/January with pod maturation. Flowering generally occurs in September before the start of the rains but sometimes high winds, late frosts or unusual early rains adversely affect flower fertilization and subsequent pod production.

A randomized complete block design was used with 8 blocks and a single tree per block. A 10 m × 10 m spacing was selected since this was predicted to be the closest spacing necessary to obtain commercial lumber production (Felker et al., 1990) i.e. 40 cm diameter trunks. The codes for the clones were for the family, block and tree of the field position where they were cloned. Thus there were two individuals represented from family 5, family 6 and family 12. Eight replications were used for all 12 clones except for F4B2T3 as we only had one of each clone. pods were harvested and the variability to be less in the clones would be expected from clones, rather than obligately outcrossed species, there is a tendency for the variability to be less in the clones than in the check. As can be seen in Fig. 1, the 95% CI is about half the mean for the check variety, while in some of high producing clones the 95% CI is about a quarter of the mean.

3. Results

The mean pod production per tree and associated 95% confidence intervals are shown in Fig. 1. Two of the clones had lower production in the 7th year’s growth than the control, some of them were slightly higher, and some of the clones had almost double the yield of the control i.e. F5B1T4, F6B1T3, F6B7T4, F9B5T2, F12B6T1, and F5B21T3. Second in importance is that in the low pod producing year of 2007 (for the control and some clones), three clones F5B21T23, F5B1T4 and F6B1T3 maintained high pod production. As would be expected from clones, rather than obligately outcrossed species, there is a tendency for the variability to be less in the clones than in the check. As can be seen in Fig. 1, the 95% CI is about half the mean for the check variety, while in some of high producing clones the 95% CI is about a quarter of the mean.
Fig. 1. *P. alba* pod production per tree (kg) for various clones and check on a 10 m × 10 m spacing in Fernandez, Argentina for years from 2004 to 2008. Trees were planted in January 2002.

From the original 57 half sib families in the original trial, we only selected 9 families for these 12 clones. Four of these families and the check were from the Rio Dulce (sweet river) Irrigation zone of the province of Santiago del Estero and one of them was from the Pinto area of Santiago del Estero. This trial was centrally located in this Rio Dulce area. The most highly productive families were from the Castelli area in the Province of the Chaco, and Ibarreta in the Province of Formosa that are respectively about 500 and 700 km northeast in a higher rainfall area. In the original trial there were 4 half sib families from Ibarreta, 4 from Castelli, 11 from the Rio Dulce Irrigation district, and 6 from Pinto. Thus in spite of their low frequency occurrence in the trial, together the northern families from Ibarreta and Castelli had 7 of 12 elite clones and they had a tendency for greater pod production than the families from the Rio Dulce Irrigation district that had a much higher number of entries.

The pod flavor analyses revealed than none of the trees produced pods with a bitter disagreeable taste or with pods that lacked any flavor. Four of the varieties were ranked very sweet, 7 (including the check) were sweet and 4 slightly sweet. Casual observations noted great variability among the percent of mesocarp (the portion between the seeds and the pod exterior)/pod weight of the varieties. An example of this variability can be seen in Fig. 2. Clone F11B6T4 had the thickest pod and the greatest mesocarp percentage. The thickness of the pod of clone F4B2T3 was very similar to that of clone F5B6T2. However in clone F4B2T3, the horizontally located endocarp containing the seed (perpendicular to the plane of the photo) is much less obvious than in clone F5B6T2 due to the

Fig. 2. Variation in mesocarp percentage of *P. alba* pods for clones F11B6T4 which had the greatest mesocarp percentage, clone F4B2T3 which had a medium mesocarp percentage and F5B6T2 which had the least mesocarp percentage. The locations of seeds, which are arranged perpendicular to the long axis of the pods, are less visible in pods which have a thicker mesocarp. The ruler is in cm.
presence of a greater mesocarp percentage in clone F4B2T3. It is the mesocarp that is important in the milling process to make human food from *Prosopis* pods (Felker et al., 2003). There is a need for a food science study to examine the percentages of pod fractions and their chemical compositions. Preliminary work comparing *P. alba* pods to the very bitter *P. juliflora* pods from Africa indicates that due to much higher citric acid in the latter pods, the acid/sugar ratio in *P. juliflora* is nearly 5 times higher than for *P. alba* (Takeoka and Felker, 2009, unpub obs).

The trunk diameter, height and canopy diameters of the clones are presented in Table 1. As might be expected for this broad canopy *P. alba*, in all cases the canopy diameter was greater than the height. The canopy diameter ranged from 6.3 to 8.3 m and thus was well over 50% of the 10 m spacing for this plantation. In all likelihood canopy closure will occur prior to the 10th year. In contrast to the increase in pod production of the clones over the control, the height, canopy diameter and trunk diameter of the clones were not greater than the control. In fact the check had greater diameter, the second greatest height and was in the upper tier for canopy diameter.

One possible explanation for the lower biomass of the clones could be due to the root system of the clones. We have noted that the root system of the cuttings is more fibrous without a dominant tap root (Felker and Ewens, unpub obs). In support of this supposition, none of the check trees have blown over in heavy wind storms, while a considerable number of the clones rooted from cuttings have blown over. In Fig. 3 we have plotted the 5-year cumulative pod production versus tree biomass of the clones and the check. The clones seem to fall along a line with the check being an outlier with higher biomass per tree and lower pod production. This would seem to indicate that the clones partition a greater portion of their growth into reproduction rather than productive tissue. However in greenhouse studies Wojtusik and Felker (1993) found a pronounced effect of rootstock on above ground biomass of various *Prosopis* species.

We have previously reported mini grafting techniques for 2 mm diameter seedlings that could be used to provide a tap rooted rootstock for these clones (Ewens and Felker, 2003). Seedlings for these rootstocks could be obtained from a companion clonal seed orchard of 22 seedlings that grew in a 45 dS m$^{-1}$ hydroponic system (Velarde et al., 2003) and that are more than 5 m tall in soils whose pH values range from 9.5 to 10.3 (manuscript in preparation). A combination of a high pod producing clone with a highly salt tolerant rootstock should be an excellent combination for arid lands.

Notwithstanding the lack of improvement in height, trunk diameter and canopy growth of the clones over the check, the annual trunk diameter increases were impressive in ranging from 2.5 to 4.1 cm year$^{-1}$. For the *P. alba* clone B2V50 growing under rainfed conditions in Texas, Felker et al. (1990) found the annual diameter growth for 3-year-old trees to be 2.57 cm. Felker and Guevara (2003) examined the influence of various genetic and management strategies on the internal rate of return (IRR) of *P. alba* plantations. This economic model was based on the times from planting to harvest and costs to achieve 100 trees ha$^{-1}$ with 40 cm diameters containing 15.4 m$^{-3}$ of sawn lumber per hectare with a value of $800 per cubic meter. The model assumed no value for the pods and a different paradigm in that the landowner would contract to convert the logs to sawn lumber. The corresponding internal rates of return for various years to harvest and annual diameter growth rates were; 10% for 24 years and 1.67 cm year$^{-1}$, 16.3% for 17 year and 2.35 cm year$^{-1}$, 18.7% for 15 year and 2.67 cm year$^{-1}$, and 27.8% for 11 year and 3.63 cm year$^{-1}$. Thus excluding the value for pod sales, based on the 2.5–4.1 cm diameter growth rates (Table 1) it would seem that the internal rate of return for this system would be about 20%.

This first comparison of *P. alba* clones in a replicated field trial also revealed significant differences in resistance to fungal pathogens, insect resistance and form. For example, clone F9B5T2 from Castelli has not suffered any attack from Psyllid insects while clone F6B7T4 from Ibarreta and F52B1T3 from Pinto are very susceptible. With respect to fungal diseases, in the fall clone F8B7T4 from Castelli has not suffered any attack from Psyllid insects while clone F6B7T4 and F17B2T2 also from Río Dulce are very susceptible. With respect to fungal diseases, in the fall clone F8B7T4 from Castelli is quite erect while clone
P. juliflora from Río Dulce has a broad spreading canopy. Three years of crossing between resistant and susceptible clones and erect versus broad canopy clones have produced several hundred seeds that have been germinated for a mapping population to study segregation for these characters in P. alba. Ranking and analyses of the percentages of progeny for disease resistance should help in identifying the number of genes responsible for this resistance. This in combination with new molecular tools for identifying disease resistance genes using degenerate primers based on conserved domains of disease resistance (Sharma et al., 2009) should greatly aid in the development of new disease resistant Prosopis high pod producing clones.

It is interesting that while the same genera of insects and fungal pathogens have evolved to attack North and South American native Prosopis species, the resistance of the Prosopis species to the pathogens/insects appear to be complementary. For example, while Pestalotiopsis guepinii (Desm.) Stey. caused complete mortality to P. glandulosa var. torreyana from the low humidity deserts of California, it had little effect on P. alba from Santiago del Estero growing in the same field trials in Texas (Lesney and Felker, 1995). However in these Argentinean trials, a Pestalotiopsis spp (evidently of another species) defoliated P. alba. Similarly while Psyllid insects (Alphalaroida sp) caused very great damage to the California native P. glandulosa var. torreyana in a California field trial (Felker et al., 1981), P. alba from Santiago del Estero experienced very little damage. However in these Argentinean trials, psyllids were the major insect pest. Thus a fungal pathogen of the genus Pestalotiopsis and Psyllid insects have co-evolved in north and South America to attack P. glandulosa var. torreyana and P. alba respectively. However the Prosopis from South America are not susceptible to the pathogens/insects from the same genera in North America. It remains to be seen if the reverse is true.

4. Discussion

In our previous work (Felker et al., 2001) we made 12 individual tree selections based on superior biomass production, pod production and pod flavor from the 1289 trees in the trial. Due to edaphic-genetic interactions it was not certain if these 12 trees were really superior. From this replicated trial with 5 years data, it is clear that some of the clones are superior for pod production. In a previous paper we (Felker et al., 2001) had calculated a narrow sense heritability of 0.244 for pod production. Odool et al. (1986) have calculated intraclass correlations which estimate the degree of resemblance among family members for Prosopis pod protein and sugar content. Across 3 sites and 3 irrigation treatments, these correlations ranged from 0.30 to 0.40 for sugar content and from 0.04 to 0.60 for protein content, indicating a generally high degree of genetic control for these families in the environments tested.

There is considerable room for improvement in the taste profile of P. juliflora that is native to Haiti and widely naturalized in Sahelian Africa, the Middle East, India and Pakistan. While sensory characteristics of P. juliflora were not compared to P. alba in our Argentinean trials (Felker et al., 2001) or to P. pallida in our Peruvian trials (Alban et al., 2002), from our experience with P. juliflora in field trials in Haiti (Lee et al., 1992) and India (Harsh et al., 1996) and casual experience in Sudan, Yemen and Kenya, the African P. juliflora would have a taste rating of very bitter on the previously described scale (Alban et al., 2002, Felker et al., 2001) of very sweet, sweet, bitter or very bitter. In P. alba and P. pallida, the mesocarp of the pods is used for human food applications due to the lack of the difficult to grind fibrous endocarp (Felker et al., 2003), the abundance of the hexanal which is an indicator rancidity in whole ground pods (Takeoka et al., 2008), and the greater concentration of the major volatile, 5,6-dihydrro-6-propyl-2H-pyran-2-one, with a coconut-like mixed with a peppermint aroma in the flours made from the mesocarp (Takeoka et al., 2008, 2009). Nevertheless by diluting the entire ground pods of P. juliflora with 4 volumes of wheat flour, acceptable baked products were made in Kenya (Choge et al., 2007). Approximately 45% of the pod mass can be made into a flour for human food use with the rest of the pod being suitable for use for animal feed.

The 50 kg per tree pod yield obtained by clones F5B1T4, F6B1T3 and F6B7T4 in the 7th growing season corresponds to 5000 kg ha⁻¹ at this 10 m × 10 m spacing. This yield compares favorably to other grains such as sorghum or wheat that can be grown at this annual rainfall of 678 mm (plus 100 mm irrigation). For example in Tanzania with 560 mm rainfall and various combinations of fertilizer (up to 150 kg N ha⁻¹), manure and pigeon pea–maize rotations, the maize yields did not surpass 3 Mg ha⁻¹ (Kimaro et al., 2009). While the rainfall plus irrigation was lower in Tanzania than Argentina, the yields were both in the 3–5 Mg ha⁻¹ range. In the adjacent Province of the Chaco, Argentina where the rainfalls are higher than in the Province of Santiago del Estero, maize yields ranged from about 1000 to 4000 kg ha⁻¹ from the period from 1969 to 2008, http://www.siia.gov.ar/sst_pacias/consultab_pcia.php. This Prosopis pod yield was also obtained without N fertilization and on soils with an EC of 8.6 dS m⁻¹ and a pH of 7.7 for the 0–30 cm depth, which would not be suitable for grains such as wheat, maize, or sorghum.

The 10 m × 10 m spacing of this trial was selected since this is the minimum spacing possible to achieve a 40 cm diameter trunk (minimum size required for a hardwood sawmill) in semi-arid regions (Felker et al., 1990). However since this plantation was established, we have found that closer spacings at earlier ages promotes a more erect, less “open grown” canopy, and that by bending the flexible stems vertically, more erect trunks can be obtained that would provide better quality lumber. Research is needed into more dense earlier spacings, that would be later thinned to 10 m spacings, in order to maximize pod production per ha and form for lumber.

The much greater pod production in the 7th year of this trial (ca 50 kg/tree) over the 1.75 kg pod/tree in the 8th year of the progeny trial (Felker et al., 2001) is due to the much closer spacing in the progeny trial (4 m × 4 m) and the fact that this trial had a very low level of management with weeds and shrubs over 1 m tall.

It is to be cautioned that evidently due to the long dry season, P. alba is not as well adapted to the true trampic as is P. pallida. For example in field trials in Haiti (Lee et al., 1992; Wojtusik and Felker, 1993), Cape Verde (Harris et al., 1996) and India (Harsh et al., 1996), P. pallida from Peru had greater height growth and was also thornless with sweet palatable pods. Nearly thornless, erect fast growing P. pallida with high production of sweet pods have been cloned (Alban et al., 2002) but these clones have not been evaluated in replicated trials.

In summary, this first replicated trial of multipurpose P. alba clones has clearly demonstrated that very large increases in pod production are possible with clones. The pod production in the 7th growing season was on a par with many other semi-arid land crops with less fertilizer input but with intensive insect control measures. Due to genetic sources of insect resistance among the clones, it would appear that insecticide uses and costs can be reduced with clones selected for insect resistance. The clones did not improve biomass production, height or diameter growth but possibly by grafting these scions onto highly saline and sodic tolerant rootstock this can be improved. Even without improved biomass production, with good management as carried out in this trial, internal rates of return for lumber production of approximately 20% may be possible.
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References


