8. Conclusions

During 2001-2004 field seasons, a total of 300 plants of *P. santalinus* and 30 plants each of *R. serpentina* and *R. tetraphylla* were collected from various locations of Andhra Pradesh and are being grown in the experimental field, field gene bank and seeds were stored in the seed bank. Seeds of *P. santalinus* collected from Papireddypally and Balpally (Kadapa District) were found to be of superior quality, in comparision to seeds from other six locations based on *in vivo* and *in vitro* germination studies.

*In vitro* regeneration studies of *P. santalinus* revealed that seeds collected from Balpally showed maximum number of multiple shoots. Since the percentage survival was very low this protocol cannot be recommended for *ex situ* conservation. This protocol is genotype and location dependent (Balpally) unlike the seeds collected from any other locations. Seed germination and *in vitro* regeneration studies indicated that Balpally area in Kadapa District could be considered as the reservoir of elite germplasm.

Distinct morphological variations were observed in *P. santalinus* pod characteristics (weight, width, and length), leaf characteristics (length, width), shoot length and number of nodes in five months old germinated seedlings collected from 16 different locations, which could be due to environmental and edaphic factors.

The wide variation in genetic distance among the accessions of *P. santalinus; R. serpentina* and *R. tetraphylla* revealed by RAPD markers reflected a high level of DNA polymorphism due to outcrossing.
"P. santalinus" is endemic to Kadapa (Cuddapah), Nellore, Chittoor and Prakasam Districts of A.P. Endemics are generally reported to have low levels of genetic variation. However, in the presently investigated accessions, 100% polymorphism was observed. Therefore, possibly the Kadapa (Cuddapah) district of A.P might be the center of diversity for "P. santalinus".

The presence of many unique markers in "P. santalinus"; "R. serpentina" and a few in "R. tetraphylla" may be due to the relatively high rate of mutations in RAPD loci. Such markers are important, as they may be diagnostic for particular regions of the genome and are accession specific.

"P. santalinus" and "R. serpentina" exhibited high genetic polymorphism. Based on the observations of this study and cluster analysis it can be suggested that for "P. santalinus" the in situ conservation measures should include Balpally (Kadapa District) and Tirupathi (Chittoor District) and for "R. serpentina" Sukumamidi (Khammam District) and Araku (Vishakapatnam District). Seeds of these sites need to be conserved (ex situ). Further sampling covering wider geographical areas, covering larger population and using more number of RAPD primers would be useful for diversity analysis.

The presence of similar markers between "R. serpentina" and "R. tetraphylla" indicates the possible intragenomic recombination or translocations. These observations are important to explore the possibility of transferring genes of agronomic and commercial importance from "R. serpentina" (for eg. reserpine) to "R. tetraphylla" which produces abundant biomass and has wider ecological amplitude.
Considerable genetic variation was observed in Sukumamidi (Khammam District) accessions of *R. serpentina* important for conservation and further progeny trials. However accessions from Dulapally (Medak District) exhibited monomorphism (genetically homogenous).