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RESEARCH ARTICLE

Variation in leaf morphological traits of *Terminalia arjuna* Roxb. in natural population of lower parts of Achanakmar Amarkantak Biosphere Reserve (AABR) of Central India.

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Abstract

Terminalia arjuna (*T. arjuna*) is a one of the important forest tree species of Achnakmar Amarkantak Biosphere Reserve (AABR) of Central India. *T. arjuna* is regarded as a potent medicinal plants for cardiac disease. A little information is available about leaf trait variations along the AABR forests of Central India. The present study on the morphological variation has been done in three natural populations of *T.arjuna* (Achanakmar, Chhaparwa and Lamini) growing in the forests of AABR. Leaves of tree species can be good morphological markers to analyse the diversity status of a forest tree species. Eleven leaf morphological traits as leaf length, leaf width, petiole length, leaf area, leaf fresh weight, leaf dry weight, leaf thickness, distance between veins, length of midrib, number of secondary veins and length of veins were measured in groups of population. In the present study the results showed that all measured leaf morphological traits have remarkable phenotypic variability. The morphological dissimilarities of the leaf in *T.arjuna* in AABR are possibly attributed to its genetic variations, developed as a result of adaptation to diverse environmental conditions. Correlation matrix among different leaf morphological traits of *T. arjuna* for different locations of AABR have been done. A positive correlations were observed in different morphological traits of leaves of *T. arjuna.* However multisite phenotypic analysis are required in order to completely separate environmental and genetic factors explaining the observed level of natural variability in this forest tree species.

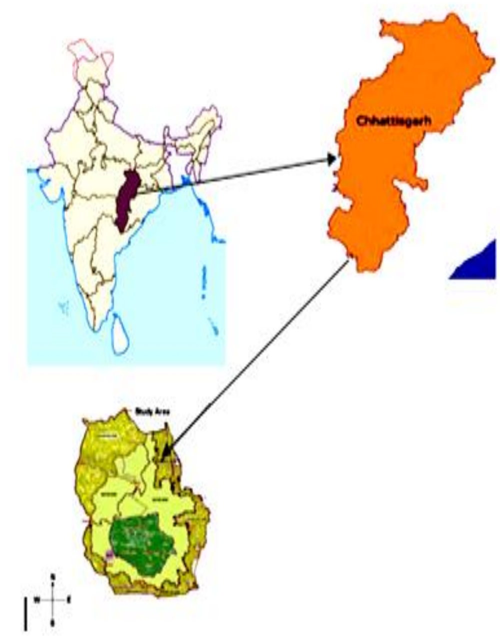
Introduction:-

Achanakmar Amarkantak Biosphere Reserve (AABR) Chhattisgarh, India is a biodiversity hub in Central India. AABR is very rich with high diversity of natural flora and fauna. It comprises of 1527 species of identified flora and 324 identified fauna. The rich dense forest of biosphere reserve is dominated by Sal tree (*Shorea robusta*) and its associates support a number of ecosystem services. The *Terminalia arjuna* is also an important part this natural ecosystem and is an evergreen forest tree (Family Combretaceae). The tree is regarded as the moisture indicator of the forests as it grows along the banks of natural water ways. This forest tree is potentially medicinal because every part of tree has got medicinal importance (**Sultana et.al., 2007; Maulik and Talwar, 2012**) and also plays an important role in the sericulture industry (**Orwa et.al., 2009**). The tree provides the natural health care to the tribal people living inside the Biosphere reserve. For the substantial protection and proper management for this species, we depend on our knowledge about ecological needs and genetic diversity of this valuable species. One of the oldest classification methods of plants for their diversity studies are their morphological characteristics (**Wang *et al.*, 2001**). In tree diversity analysis leaf morphology induces important phenotypic trait (**Bruschi *et al.*, 2003; M C Donald *et al.*, 2003; Kaffash *et al.*, 2008; Zarafshar *et al.*, 2009**).Leaves are highly important organs of a tree, sensitive to growth conditions, especially during a leaf expansion phase (**Masarovicova 1988, Bayramzadeh et al. 2008**).The leaves of the trees has the ability to be adapted to any site from which they originate (**Garcia-Plazaola**

& Becerril 2000, Wittmann et al. 2001, Toan et al. 2010, Amjad Ali et al. 2011) by altering the proper morphological and anatomical changes(**Bussotti et al. 2000,Gratani et al. 2003**). The leaf variations are being used effectively as an efficient morphological markers to analyse the diversity within and among different forest tree species growing in their natural habitat.The morphological traits of the leaves of the forest tree species can provide a wide range of information about the evolution, genetics and its physiology (**Main 1966**). The work on the morphological variation of different leaf traits of *T. arjuna* in Achanakmar Amarkantak Biosphere Reserve of Central India have not been done so far. The present study provides an insight about the diversity of *T. arjuna* in AABR

Material and methodology:-

Achanakmar-Amarkantak Biosphere Reserve is located in the states of Madhya Pradesh and Chhattisgarh.The reserve covers a huge area of 3835.51 sq. km. About 68.10% out of the total area of this biosphere reserve lies in the Bilaspur forest division, Bilaspur, Chhattisgarh. The core region of Achanakmar Amarkantak Biosphere Reserve falls in Chhattisgarh state lies between 22^0 15' to $22^0$58'N and 81^0 25' to 82^0 50'E The altitude varies from 400-1100 m above the mean sea level. The vegetation of the area is of subtropical type dominated mainly by Sal trees. The mean annual temperature ranges between 21° C to 31°C. The average rainfall is about 1, 900 mm which is received largely from South West monsoon. The soils of the area are usually lateritic, alluvial and black cotton type.



The fully grown trees in three different locations i.e, Achanakmar, Chhaparwa and Lamini ranges of AABR between an altitude of 300-600 m.s.l. were selected randomly and 25 trees from each location were selected for the study of morphological variation of leaves. Ten expanded leaves (sunned leaves from the middle part of tree crown) were collected from fully mature and healthy trees each, that were 30-50 cm in diameter at breast height. Leaf length (LL, mm), leaf width (LW, mm),leaf area (LA, cm^2), and distance between veins (DBV, mm), petiole length (PL) were analysed. A digital caliper with an accuracy of 0.01 mm was used to measure petiole length (PL) Leaves were oven-dried at 80º C for 72 hours and weighed for the calculation of leaf dry weight(LDW). The leaf thickness (LT, mm) was also measured on leaf cross sections.

Statistical Analysis.-

Means of the morphological traits of every population were compared using Fisher tests of ANOVA analysis at 5% and 1% levels of significance. Pearson coefficient (r) of correlation between the different morphological traits were also calculated.

Results:-

Morphological leaf traits of *T. arjuna* in the lower parts of AABR sowed a remarkable difference. The Achanakmar area had the largest leaf length (25 cm),leaf width (8cm),Fresh and dry weight (5.58 gm and 1.6 gm), leaf area (154.4 cm^2),leaf thickness (1.1mm) ,length of petiole (1.22 cm) and distance between primary veins (1.53 cm) respectively (fig: e to fig l).

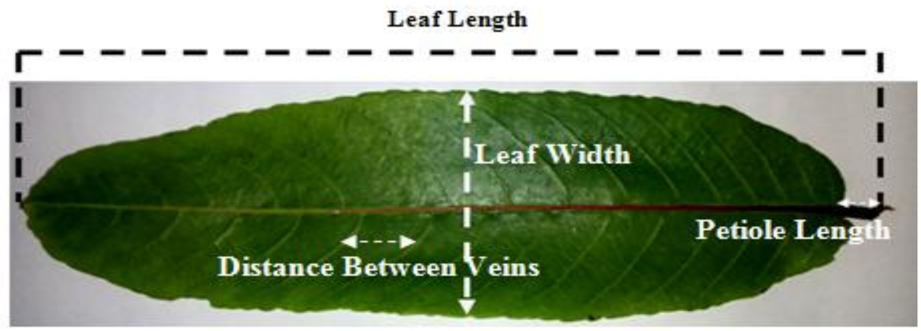


Fig a: Diagram of *T. arjuna* leaf illustrating measurements of leaf length (*LL*), petiole length (*PL*), leaf width (*LW*) and distance between veins (*DBV*)

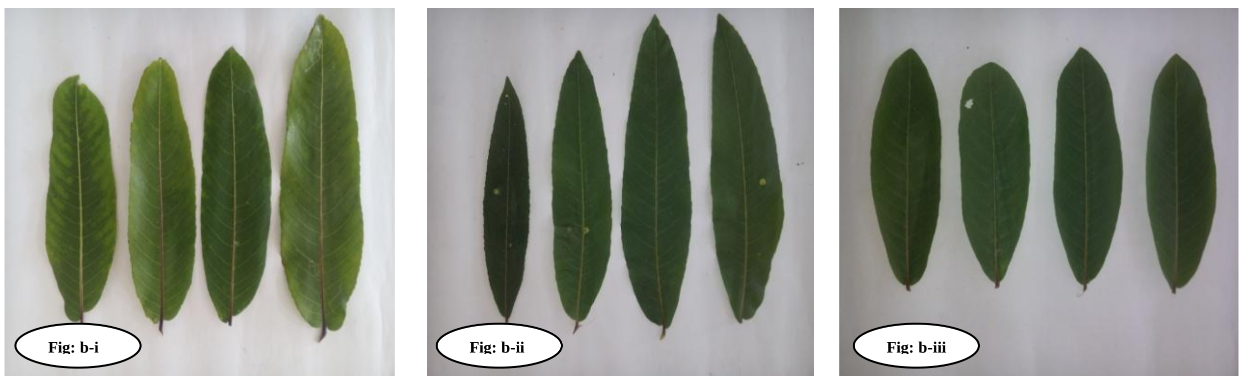


Fig: b- i, ii and iii: Showing phenotypic variation at different population sites of *Terminalia arjuna* leaves in AABR

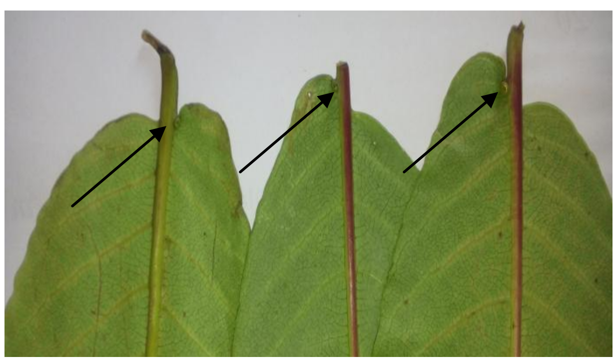


Fig c: Presence of only one leaf gland under ventral surface at Achanakmar site of AABR

Fig d: Presence of two leaf glands under ventral surface at Chhaparwa and Lamini site of AABR.

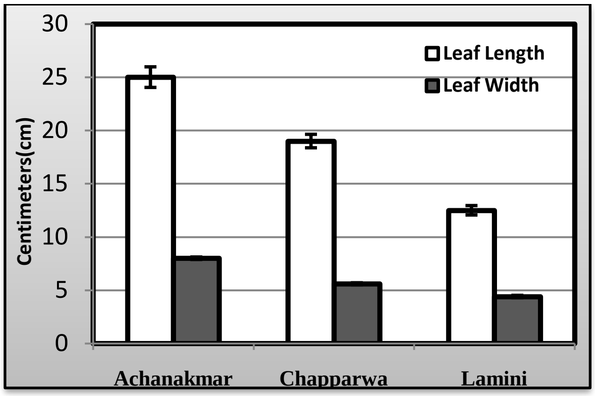


Fig e : ***Terminalia arjuna-*** The fig shows the variation of average leaf length and width in different localities of AABR.The data shown in the graph are mean ± SE of four replicates. Bars represent standard errors within treatment means. The results are statistically significant at 0.05% level of significance

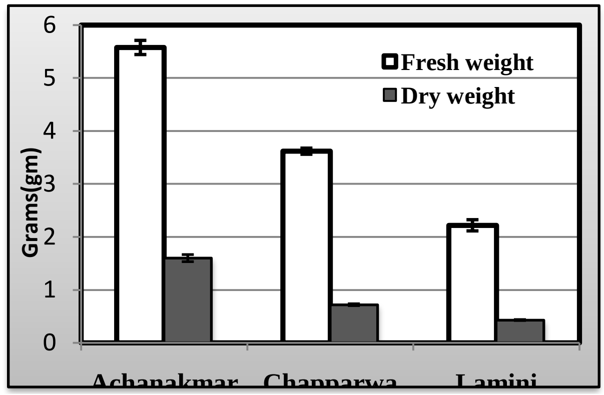


Fig f : ***Terminalia arjuna-*** The fig shows the variation of average fresh weight and dry weight of leaves in different localities of AABR.The data shown in the graph are mean ± SE of four replicates. Bars represent standard errors within treatment means. The results are statistically significant at 0.05% level of significance

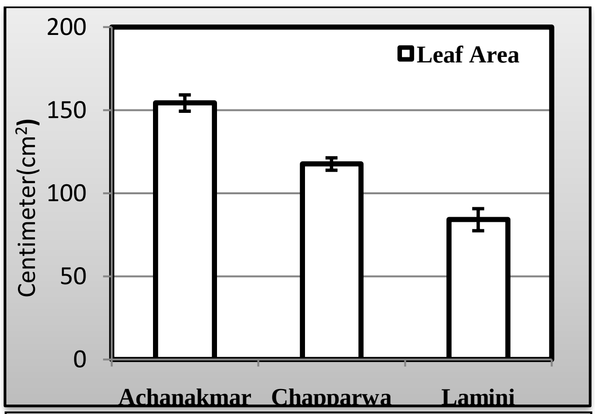


Fig g : ***Terminalia arjuna-*** The fig shows the variation in average leaf area in different localities of AABR. The data shown in the graph are mean ± SE of four replicates. Bars represent standard errors within treatment means. The results are statistically significant at 0.05% level of significance

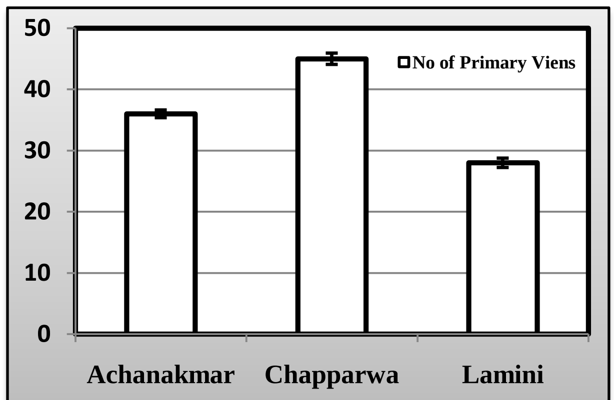


Fig h : ***Terminalia arjuna-*** The fig shows the variation in average number of veins in different localities of AABR.The data shown in the graph are mean ± SE of four replicates. Bars represent standard errors within treatment means. The results are statistically significant at 0.05% level of significance

The Lamini forests bear the smallest leaf length (12.5 cm),leaf width (4.4cm),Fresh and dry weight (2.2 gm and .43 gm), leaf area (84.2 cm^2),leaf thickness (1.1mm) ,number of primary veins (28) and length of primary vein (2.6 cm) respectively (fig e to fig l). However maximum number of primary veins were found in Chhaparwa (45) this was due to the less distance between the secondary veins (.70 cm) in the leaf of *T. arjuna*. It was found that the length of petiole in Lamini population was maximum(1.22 cm)and highest than the other two populations (fig l).

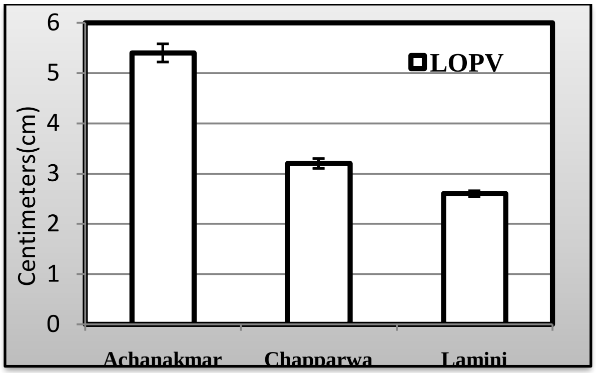


Fig i : ***Terminalia arjuna-*** The fig shows the variation in average length of primary veins(LOPV) of leaf in different localities of AABR.The data shown in the graph are mean ± SE of four replicates. Bars represent standard errors within treatment means. The results are statistically significant at 0.05% level of significance

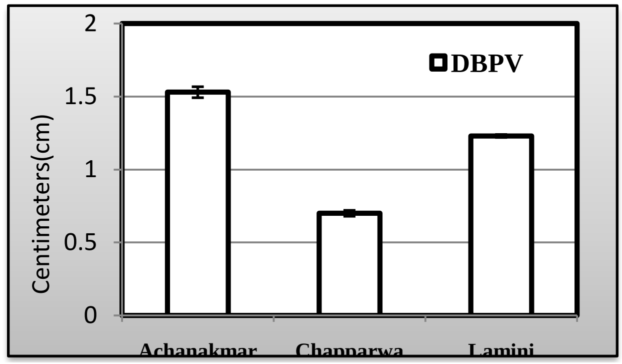


Fig j : ***Terminalia arjuna-*** The fig shows the variation in average distance between primary veins (DBPV) of leaf in different localities of AABR.The data shown in the graph are mean ± SE of four replicates. Bars represent standard errors within treatment means. The results are statistically significant at 0.05% level of significance

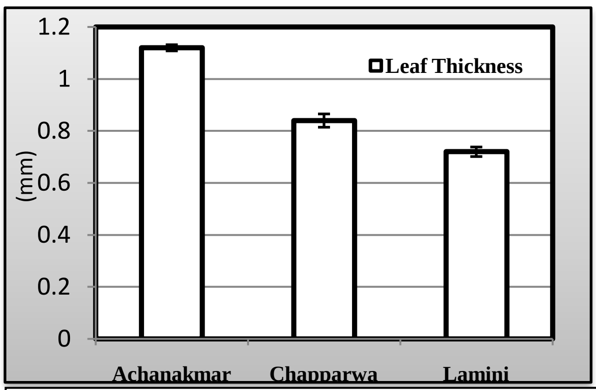


Fig k : ***Terminalia arjuna-*** The fig shows the variation in average leaf thickness in different localities of AABR.The data shown in the graph are mean ± SE of four replicates. Bars represent standard errors within treatment means. The results are statistically significant at 0.05% level of significance

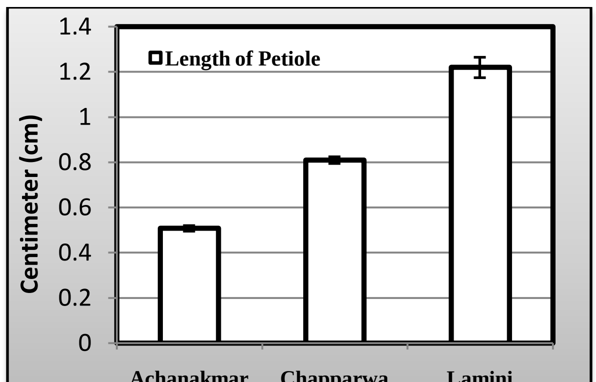


Fig l : ***Terminalia arjuna-*** The fig shows the variation in average length of petiole in different localities of AABR.The data shown in the graph are mean ± SE of four replicates. Bars represent standard errors within treatment means. The results are statistically significant at 0.05% level of significance

Leaf of *T. arjuna* has a pair of knob-like glands on the ventral (lower) side at the junction between the petiole and the lamina. The ventral surface has downy hair but the ventral surface was smooth. Variation in the number of glands found in the ventral side of leaf was observed .The leaves of Achanakmar region bear only one gland (fig-c) were as the leaves of Chhaparwa and Lamini bear two glands (fig- d).Another marked variation in the color of the midrib was observed. The leaves of Achanakmar have a red colored midrib and the leaves of Chapparwa and Lamini has green colored midrib.

Table 1: Correlation matrix among different leaf morphological traits of *Terminalia arjuna* in different localities of AABR,Chhattisgarh,India

Parameters	***LL***	***LW***	***LT***	***LA***	***LFW***	***LDW***	***PL***	***DBV***	***LMR***	***NPV***	***LPV***
LL	**1**										
LW	**0.977**	**1**									
LT	**0.968**	**0.999**	**1**								
LA	**0.998**	**0.986**	**0.979**	**1**							
LFW	**0.992**	**0.995**	**0.991**	**0.997**	**1**						
LDW	**0.953**	**0.995**	**0.998**	**0.967**	**0.982**	**1**					
PL	**0.968**	**0.999**	**1**	**0.979**	**0.991**	**0.998**	**1**				
DBV	**0.335**	**0.527**	**0.557**	**0.381**	**0.444**	**0.603**	**0.557**	**1**			
LMR	**0.999**	**0.974**	**0.965**	**0.998**	**0.991**	**0.949**	**0.965**	**0.323**	**1**		
NPV	**0.490**	**0.295**	**0.259**	**0.446**	**0.383**	**0.204**	**0.259**	**0.656**	**0.501**	**1**	
LPV	**0.931**	**0.987**	**0.992**	**0.948**	**0.968**	**0.997**	**0.992**	**0.654**	**0.927**	**0.140**	**1**

Pearson coefficient (*r*) of correlation between pairs of leaf morphological traits (n = 15). (*LW* – leaf width, *LL* - leaf length, *LA* - leaf area, LFW-leaf fresh weight, LDW- leaf dry weight, PL - petiole length, , *LT* - leaf thickness, *DBV* - distance between veins, *LMR*-length of midrib, *NPV*-number of primary veins,*LSV*-length of veins)

The results can further be explained by a stronger correlation between morphological traits. A very high positive correlation was observed between leaf width, leaf length, leaf area, -leaf fresh weight, leaf dry weight, petiole length, , leaf thickness, , length of midrib, ,length of veins of *T. arjuna* in AABR. However the number of secondary veins does not show a significant correlation with the other leaf traits also distance between veins was less correlated with the other leaf traits (Table-1)

Leaf length and leaf width were strongly correlated with leaf area(r=0.99 and 0.98). However the relationship between leaf dry mass with leaf length and width was also strong(r = 0.95 and 0.99).The different kinds of relationship among among different leaf traits were also reported by different authors (**Korner & Diemer 1987, Dijkstra 1990, Witkowski & Lamont 1991, Choong et al. 1992, Bayramzadeh et.al., 2012**)

Discussion:-

The morphological traits in leaves have an important role in determining the difference between trees in various habitats (**Sokal & Rohlf, 1995**).High phenotypic plasticity in leaf characters of *T. arjuna* growing in lower parts of AABR was observed. The findings of the study shed light on the diversity of *T. arjuna* in the lower parts of AABR.

The reduction of the leaf size (leaf length, leaf width and leaf area) in *T. arjuna* along an altitudinal gradient was observed. The leaf size in Achanakmar populations was more than the leaf size of Chhapparwa and Lamini populations. Reduction in leaf size (lamina length, lamina width and leaf area) along an altitudinal gradient has been reported in *Metrosideros polymorpha* Gaud. (**Cordell et al., 1998**) and *Quercus aquifolioides* (**Li et al. 2006**).This type of phenotypic plasticity in leaf size have been observed in several studies (**Meinzer et al., 1985; Korner, 1989; Havstrom et al., 1995**).The increase of the leaf size can supplement a number of ecosystem services like Increase in carbon sequestration, increase in biomass, increase in rate of evapotranspiration etc and also the large leaf size of *T. arjuna* can assist in the increase in silk production .

The length of the petiole is of pivotal importance because it determines the position of leaf blades within the canopy. The populations of Lamini site have longer petiole than the populations of Chhapparwa and Achanakmar sites. The length of the petiole causes two developmental processes, cell division and cell elongation (**Jelmer et al., 2008**). Achanakmar falls in buffer zone and the forests are not as dense as the forests of Chapparwa and Lamini which falls in core area of AABR. The trees of Lamini population grow in dense forests where the light penetration is restricted and the length of petiole is elongated. The length of petiole is more in the trees growing in shade than the trees growing in the light. The result was supported by the work done on *Trifolium repens* (**Jelmer et al., 2008**).Much is known about the molecular basis and the signal transduction pathways of shade-induced elongation responses of petiole (**Chen et al., Weijschede et al., 2006, Weijschede et al., 2008**) as well as about their ecological and evolutionary implications (**Donohue et al., 2000 ; Huber et al., 2004**)

Leaf traits are said to be correlated as one trait is effected by another trait. A positive correlation was observed between the different leaf traits of *T. arjuna* .These results were also confirmed by (**Teklehaimanot et al. 1998; Choong et al. 1992, Bayramzadeh et.al., 2012**) who have reported that the morphological traits of leaf have correlation.

Phenotypic variation in the leaf size can be due to intrinsic (genetic) factor or plastic (environmental) factor. Moreover the interplay of genotypic differences and induced plastic responses causes trees to express differences in morphological traits (**Evans and Turkington, 1988 ; Aarssen and Clauss, 1992 ; Stratton, 1995)** and causes the species to get genetically diversified.Thus a variation in the leaf characters clearly identifies the genetic diversity in *T. arjuna* in three natural populations of AABR which is a healthy sign of genetic adaptability and better gene pool dynamics.

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