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Influence of Hybridization on the Traits of Silk Production and Filament Denier in Indian Tropical Tasar Silk Insect, *Antheraea mylitta* Drury

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Abstract: The aim of the present study is to explore hybridization influence to achieve the potential of silk yield and quality in commercial tasarculture, as against the current status of applying only a few wild and semi-domesticated ecoraces of *Antheraea mylitta* D to produce raw silk during commercial crop rearing season (September-December). The four F_1 hybrid combinations generated using Daba, Jata and Raily tasar ecoraces of seed crop rearing season (July-August) were evaluated during commercial crop season of 2007 and 2008 for the influence of hybridization as positive heterosis in the traits of economically important silk productivity and filament denier. The F_1 hybrid, Daba x Jata has recorded positive heterosis in single shell weight (+25.6%), total silk yield (+79.0%) and silk filament length (+68.1%), but for silk filament of high denier (11.98 d) followed by its reciprocal hybrid, Jata×Daba in shell weight (+18.8%), silk yield (+68.1%), filament length (+63.8%) and filament of highest denier (12.32 d). Though, the Raily×Daba hybrid could show highest heterosis in shell weight (2.47 g) with filament of least denier (10.86 d), the improvement in filament length was marginal (+44.7%) and heterosis in silk yield was negative (-47.0%), while the performance of Daba×Raily hybrid was least among the hybrids. The study infers hybridization influence on silk related traits at F_1 level as positive heterosis and the application feasibility of Daba×Jata and Jata×Daba hybrids during commercial crop rearing season for higher silk productivity with silk filament denier at least better than one of the parents of F_1 hybrid combination.

Key words: *Antheraea mylitta*, F_1 hybrids, heterosis, silk filament denier, silk yield traits

INTRODUCTION

The tropical tasar silk is produced by the wild sericigenous and polyphagous insect, *Antheraea mylitta* Drury (Lepidoptera: Saturniidae), distributed all along central India (12-31° N latitude and 72-96° E longitude). Though, the insect species has rich genetic resource with forty four races on acclimatization to diverse ecozones, only the Daba and Sukinda races are semi-domesticated and applied for commercial rearings and their hybrid vigour is yet to explore (Hansda *et al*., 2008; Ojha *et al*., 2009). The inclusion of silkworm parents of different eco-geographical origin in breeding leads to improvement of traits associated with silk (Petkov *et al*., 2000; Reddy *et al*., 2009a). The genetic interaction through

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hybridization not only provides mid and better parent heterosis (relative heterosis and heterobeltiosis) but also contributes to improved silk quality due to the genetic diversity of the parental races involved in the breeding process (Umadevi *et al*., 2005; Reddy *et al*., 2008, 2009a). The fecundity, egg hatching, cocoon yield and shell weight will together contributes for total silk yield (Reddy *et al*., 2010b). The existing genetic variation among wild and domesticated silk insects needs exploitation through hybridization for yield gain in terms of quantity and quality, as the introgressive hybridization with parental diversity has heterosis impact on silk quality with cumulative effect of genes (Verma *et al*., 2003; Reddy *et al*., 2008, 2009a, c; Seshagiri *et al*., 2009). The heterosis in respect of cocoon weight, shell weight, filament length, denier and rendita correlates with parental genetic divergence (Mirhoseini *et al*., 2004; Talebi and Subramanya, 2009). The potential phenotypic expression of the genotype depends on interaction with the environment (Hansda *et al*., 2008; Ojha *et al*., 2009; Reddy, 2010), as the gene being endogenic factor play major role and environment being the exogenic factor influences the phenotypic expressivity (Moorthy *et al*., 2007; Reddy *et al*., 2010a). The extent of hybrid vigor in silk related traits like filament size deviation reported varying under different temperatures, nutrient availability, feeding duration and crowding as they influence the insect body plasticity (Kumar *et al*., 2003; Davidowitz *et al*., 2004; Malik and Reddy, 2007; Zhao *et al*., 2007; Reddy *et al*., 2009b, d). The high productivity and narrow adaptability to fluctuating environment of *Bombyx mori* indicates the genetic stability of breed (Gangwar *et al*., 2009). The silkworm hybrids in general record better reeling performance over pure races (Takabayashi *et al*., 1994; Rao *et al*., 2004) and the correlation among shell weight and filament length was positive, while between filament length and denier it was negative (Sekharappa *et al*., 1999; Radhakrishna *et al*., 2001; Petkov *et al*., 2003; Doddaswamy *et al*., 2009). The limitation of breeds in tasar silkworm requires the combining of productivity and quality of silk made the authors to choose three diverse ecoraces, i.e., Daba of Jharkhand, Jata of Orissa and Raily of Chhattisgarh regions of India in the present study.

MATERIALS AND METHODS

The three parental ecoraces Daba, Jata and Raily of tropical tasar silkworm, *A. mylitta* belongs to different eco-geographic origin were initially reared in a Randomized Block Design (RBD) with three replications each during the seed crop rearing season (July-August) of 2007 and 2008 on the economic plantation of *Terminalia tomentosa* (W and A) maintained at field laboratory of Central Tasar Research and Training Institute (CTR and TI), Ranchi, India by following the integrated package of tasar silkworm rearing. The generated non hibernating healthy and live seed cocoon stocks from the said rearing were preserved in the tasar grainage house following integrated package of seed cocoons preservation and conducted grainage for silkworm seed (egg) production. The fresh moths emerged during the month of September were used to produce disease free layings (Dfls) of F_1 hybrids viz., Jata×Daba, Raily x Daba and their reciprocals in addition to pure layings of parents through selfing. The three parental ecoraces along with four F_1 hybrids were reared simultaneously in a RBD with three replications each during commercial crop rearing season (September-December) of 2007 and 2008 on the economic plantation of *T. tomentosa* at field laboratory of CTR andTI, Ranchi, following integrated package of tasar silkworm rearing. The larvae of one Dfl of parents and F_1 hybrids were considered as one replication during the grainage and crop rearings for recording the observations. The single shell weight was calculated based on twenty random cocoon samples and the silk yield was calculated multiplying the average single shell weight with the number of cocoons harvested per Dfl. The silk related technological characters like silk filament length and denier were studied on single cocoon basis with ten random samples at post cocoon technology division of CTR and TI, Ranchi. The data so obtained on the traits of silk productivity and quality were subjected to statistical analysis.

RESULTS

In situ Performance of Parental Ecoraces

The data on particulars and performance levels of parental ecoraces Daba, Jata and Raily under *in situ* habitation (Table 1) indicate variations in their place of origin, food plants, voltinism, single shell weight, absolute silk yield, length and denier of silk filament. As like their origin from different eco-geographic areas of the country, i.e., Daba from Singhbhum of Jharkhand, Jata from Simlipal of Orissa and Raily from Bastar of Chhattisgarh states in India and the ecoraces have wide variations in their commercial traits. The Jata and Raily wild races are superior in shell weight and filament lengths, while their total silk yield was low and the silk filament was thicker with high denier. The varied voltinism among the parents indicates their commercial application value and the trivoltinism and semi-domestication of Daba ecorace looks highly compatible among the parents. However, the semi domesticated Daba ecorace, though recorded lesser single shell weight, it has shown higher total silk yield with moderate silk filament length of finer denier among the three parental ecoraces preferred for the study.

Analysis of Variance for Silk Traits

The Analysis of Variance (ANOVA) of four silk interrelated commercial characters among the parents, F_1 hybrids and parents versus F_1 hybrids of commercial crop rearing season (Table 2) has revealed significant variance. The variance among the parents was recorded significant in all the silk related traits except for single shell weight. However, the variance among the F_1 hybrids was significant for the total silk yield and length of silk filament, while the single shell weight and silk filament denier were non-significant. The variance in parents versus F_1 hybrids, the measure of hybridization was found significant for all the four silk associated traits studied indicate the impact of hybrid vigour and influence on silk production as single shell weight, total silk yield and length of silk filament and quality as silk filament denier.

Table 1: Performance levels of parental ecoraces (Daba, Jata and Raily) under *in situ* habitat

				Parameters			
Parental ecoraces	Place of origin	Primary food plant(s)	Voltinism	Single shell weight (g)	Total silk yield/ laying (g)	Silk filament length (m)	Silk filament denier (d)
Daba							
Range	Singhbhum	*Terminalia*	Bi and	1.25-2.36	62.5-177.0	475-1240	9-11
Average	(Jharkhand state)	species	Trivoltine	1.80	108.0	962	10
Jata							
Range	Simlipal	*Terminalia*	Uni and	1.60-2.34	24.0-44.5	840-1550	11-13
Average	(Orissa state)	species	Bivoltine	2.05	34.85	1184	12
Raily							
Range	Bastar	*Shorea*	Univoltine	1.97-2.82	35.5-70.5	570-1750	12-14
Average	(Chhattisgarh state)	*robusta*		2.23	46.83	1385	13

Table 2: ANOVA for silk related traits among parents and F_1 hybrids of commercial crop season

		Mean sum of squares			
Source	df	Single shell weight (g)	Total silk yield (g)	Silk filament length (m)	Silk filament denier (d)
Replicates	2	0.02	82.8	12322.7	0.002
Parents	2	0.13	2982.8	139176	5.96
		NS	***	**	***
Hybrids	3	0.10	13866.9	289249	1.33
		NS	***	***	NS
Parents vs. hybrids	1	0.94	3467.5	899655	2.32
		**	***	***	*
Error (A)	12	0.06	138.6	14107	0.45
Total	11	0.18	4263.4	178302	1.06

*Significant at 5%; **Significant at 1%; ***Significant at 0.1%; NS: Non significant

Table 3: Performance levels of silk related traits of parents of seed and commercial crop seasons

Parent and crop season	Single shell weight (g)	Total silk yield (g)	Silk filament length (m)	Silk filament denier (d)
Daba seed crop	1.43±0.07	94.2±9.3	799±86	10.61±0.17
Jata seed crop	1.63±0.12	70.7±3.0	1016±79	12.59±0.36
Raily seed crop	1.75±0.07	39.3±3.2	1157±83	13.65±0.39
Daba commercial crop	1.70±0.02	95.6±6.5	880±19	11.04±0.34
	(+18.9)	(+1.48)	(+10.1)	(+4.10)
Jata commercial crop	1.82±0.12	85.6±5.3	1249±89	12.64±0.30
	(+11.6)	(+21.1)	(+22.9)	(+0.40)
Raily commercial crop	2.11±0.09	36.7±3.5	1257±56	13.85±0.46
	(+20.6)	(-6.60)	(+8.60)	(+1.50)

Values are Mean±SE, + or - percent change over the respective parents of seed crop

Performance of Parental Ecoraces During Seed and Commercial Crop Rearing Seasons

The average performance levels of parental ecoraces based on pooled data of two years (2007-2008) in respect of silk related traits during seed and commercial crop rearing seasons (Table 3) indicate similar trend by the ecoraces in both the seasons, except for higher levels of performance during commercial crop season and lesser total silk yield in Raily ecorace over its respective parent of seed crop season. However, the performance levels in respect of silk yield traits during the commercial rearing season found varied among the parental ecoraces with highest shell weight of 2.11 g and filament length of 1257 m in Raily followed by Jata and Daba with 1.82 and 1.70 g of shell weight and 1249 and 880 m of filament length, respectively. In contrary, the total silk yield was highest in Daba with 95.6 g followed by Jata and Raily with 85.6 and 36.7 g, respectively. Alike the yield parameters (shell weight and silk yield), the silk filament denier indicates the quality of silk was highest in Raily with 13.85 d followed by Jata and Daba with 12.64 and 11.04 d, respectively suggests the generation of thicker filament by Raily and Jata ecoraces than semi-domesticated Daba ecorace.

Performance of F_1 Hybrids During Commercial Crop Rearing Season

The performance levels of four F_1 hybrids on silk yield traits and filament denier during commercial crop rearing season (Table 4) revealed positive mid parent (relative) heterosis in all the traits with marginally higher filament denier in respect of the F_1 hybrid Daba×Jata and its reciprocal. However, the Daba×Raily and its reciprocal F_1 hybrids have recorded positive mid parent heterosis only in shell weight and filament length while the total silk yield and silk filament deniers recoded negative heterosis over the mid parent value. Among the F_1 hybrids, the Daba×Jata has recorded highest levels of overall performance with 2.21 g shell weight, 162.2 g total silk yield and 1790 m of silk filament length, which were at 25.6, 79.0 and 68.1% of mid parent heterosis. However, the silk filament of this hybrid was marginally thicker

Table 4: Performance levels of silk related traits of F_1 hybrids of commercial crop season

F_1 hybrid combination	Single shell weight (g)	Total silk yield (g)	Silk filament length (m)	Silk filament denier (d)
Daba×Jata	2.21±0.18	162.2±10.4	1790±129	11.98±0.25
	(+25.6)	(+79.0)	(+68.1)	(+1.2)
Jata×Daba	2.09±0.14	152.3±9.9	1744±49	12.32±0.17
	(+18.8)	(+68.1)	(+63.8)	(+4.1)
Daba×Raily	2.45±0.20	44.9±2.8	1109±19	12.18±0.49
	(+28.3)	(-32.1)	(+3.8)	(-2.1)
Raily×Daba	2.47±0.12	35.0±3.0	1546±40	10.86±0.40
	(+29.3)	(-47.0)	(+44.7)	(-12.7)

Values are Mean±SE, + or - are the mid parent heterosis (%) over parents of commercial crop

(11.98 d), with 1.2% of positive mid parent heterosis. The performance levels of Jata×Daba hybrid was next to Daba×Jata with 2.09 g shell weight, 152.3 g total silk yield and 1744 m of silk filament length, which were at 18.8, 68.1 and 63.8% of mid parent heterosis. Also, the silk filament of this hybrid was little thicker (12.32 d), with 4.1% of positive mid parent heterosis. Though, the Raily×Daba hybrid has recorded highest single shell weight of 2.47 g with an improvement of 29.3% (positive heterosis), moderate filament length of 1546 m and least denier of 10.86 d, its total silk yield was lowest (35.0 g); while the Daba×Raily hybrid has recorded same trend like Raily×Daba with marginally lower levels of performance in all the silk associated traits.

DISCUSSION

The performance levels of the parental ecoraces clearly indicate their distinctiveness and genetic diversity as like their origin from different ecozones (Daba from Jharkhand, Jata from Orissa and Raily from Chhattisgarh) of the country by feeding on different food-plants are in accordance with the earlier reports of Hansda *et al*. (2008), Ojha *et al*. (2009) and Reddy *et al*. (2009a, 2010b). The highest silk yield, finer filament denier of Daba ecorace and wider adoptability with bi and trivoltinism, in spite of lower shell weight and silk filament length prove its commercial applicability and economic viability. In contrary, the other two wild ecoraces, Jata and Raily in spite of having better shell weight and longer filament length, their lesser total silk yield, thicker filament with high denier and low adoptability with uni and bivoltinism made them comparatively less sustainable for exploitation through commercial rearings. The mixing of commercially essential traits available with both domesticated and wild ecoraces for hybridization impact at F_1 level as was reported by Moorthy *et al*. (2007), Reddy *et al*. (2009b) and Talebi and Subramanya (2009) to get a viable breed for sustainable tasarculture made to choose three assorted ecoraces (Daba, Jata and Raily) of tasar silkworm, *A. mylitta* as parents.

The significant variance among the parents in all the silk related traits except for single shell weight of commercial crop season confirms the existence of genetic divergence among the parents. The variance among F_1 hybrids however was significant only in respect of total silk yield and silk filament length suggests the higher impact of hybridization on those traits than the shell weight and silk filament denier, which were non-significant. But, the variation significance in relation to the performance of parents and hybrids, in spite of their unvarying rearing practices and environment indicates the phenotypic expressiveness of varied genotypes as parents and F_1 Hybrids. These results are corroborating with the findings of Kumar *et al*. (2003), Davidowitz *et al*. (2004), Mirhoseini *et al*. (2004) and Umadevi *et al*. (2005). The significant variance among the parents versus F_1 hybrids during commercial season indicates the varied hybridization influence as relative heterosis in F_1 hybrids in the economic traits of silk productivity as shell weight, silk yield and filament length and silk quality as denier of filament. These results are in conformity with the findings of Takabayashi *et al.* (1994), Rao *et al.* (2004) and Reddy *et al.* (2009c).

The commercial sustenance of tasarculture depends mainly on the quantity and to some extent on the quality of the raw silk produced, which normally influenced by the breed, feed and the rearing environment. The tasar silkworm rearing being an outdoor practice mostly on the nature grown food-plants, the success of tasar silk production go with the silkworm breed applied for commercial rearing. Further, the reports of Hansda *et al.* (2008) and Reddy *et al.* (2009d) reveals that the generation of raw silk mainly took place under commercial crop rearing season (September-December), where the environmental conditions and quality of the feed are superior and hence the role of breed is paramount in achieving the higher quantity (yield) of silk with better quality (denier) of silk filament. The tasar silkworm is destined to undergo diapause after the commercial rearing season; it develops thicker cocoon shell to protect its pupal form combating the environmental adversities ahead with severe winter and summer while completing its life cycle. Hence, by nature the silk insect will produce higher silk during the commercial crop rearing season than seed crop season (July-August), which completes life cycle with simultaneous metamorphosis of pupa to moth. The reports of Petkov *et al.* (2000), Radhakrishna *et al.* (2001), Davidowitz *et al.* (2004), Zhao *et al.* (2007) and Reddy *et al.* (2009c) indicates that the silkworm races produces higher silk yield during the commercial crop rearing with better genotype-environment (G×E) interaction. Hence, the tasar F_1 hybrids can be exploited for positive heterosis to produce higher quantity of silk with enhanced single shell weight, total silk yield and silk filament length and better quality of silk with reduced silk filament denier.

The enhanced levels of performance by the parents of commercial crop rearing season in respect of single shell weight, total silk yield and silk filament length over their respective parents of seed crop rearing season, except for total silk yield in Raily ecorace indicate the role of rearing environment and season in varied expression of phenotypic traits of silk by the parental ecoraces corroborating the findings of Kumar *et al.* (2003), Malik and Reddy (2007), Zhao *et al.* (2007) and Reddy *et al.* (2009d, 2010a). Though, the improvement in single shell weight and silk filament length might be due congenial rearing environment, better quality of leaf of food-plant and longer larval feeding duration, while, the total silk yield might be with the combined impact of higher fecundity, better hatching and most importantly with improved Effective Rate of Rearing (ERR) and cocoon yields, as every trait has contributory role in attaining higher silk yields (Raju and Krishnamurthy, 1993; Reddy *et al.*, 2009b, d; Reddy, 2010). However, the silk yield in respect of Raily parental ecorace has reduced marginally in commercial crop season in spite of improved shell weight and filament length, might be due to low ERR and cocoon yields and racial distinctive show, away from its native niche as reported by Davidowitz *et al.* (2004) and Ojha *et al.* (2009). But, the increase in filament thickness (denier) during commercial crop rearing season is a negative point in commercial aspect and might be due to the influence of environment with G×E interaction and preparation of tasar insect for diapause than the racial character, as the trend of filament denier remains same in all parental ecoraces.

While comparing the hybridization influence as mid parent heterosis among F_1 hybrids, the Daba x Jata has revealed strong hybridization influence as improvement in all silk related traits with elevated leap in silk yield and filament length as was earlier reported by Sekharappa *et al.* (1999), Radhakrishna *et al.* (2001), Petkov *et al.* (2003), Reddy *et al.* (2009c) and Seshagiri *et al.* (2009) indicates its commercial prospective. Though, there is marginal increase in filament denier (11.98 d) over the mid parent value, it found less than one of its parents, Jata (12.64 d) of commercial crop rearing season. Further, the levels other economic traits of silk productivity like total silk yield with 162.2 g and silk filament length with 1790 m are the highest among four F_1 hybrids evaluated parallel. The F_1 hybrid Jata×Daba could also show hybridization impact and positive heterosis, but, was next to Daba×Jata with marginal decrease in silk productivity (lower shell weight, silk yield and filament length) and quality (higher filament denier). The positive performance of F_1 hybrids, Daba×Jata and Jata×Daba suggest their commercial prospective in seed cocoon expediency and attaining the optimal silk productivity and quality. The hybridization influence and positive heterosis in F_1 hybrids, though indicates the commercial advantage, it may suite for the productivity but not with the quality of silk as positive heterosis refers to thicker filament with high denier. Further, the quality of filament (denier) is more of racial character and attaining higher silk yield with lower filament denier looks intricate as the productivity and quality are negatively correlated as observed by Sekharappa *et al.* (1999), Petkov *et al.* (2000), Verma *et al.* (2003) and Reddy *et al.* (2009c, 2010a). However, attaining better silk productivity together with quality over the existing options looks rational and hence attaining higher levels in silk productivity keeping silk quality either marginally changed either way or unchanged or vice versa is a success and has commercial advantage in tropical tasarculture. The F_1 hybrid, Raily×Daba has shown influence of hybridization as highest positive heterosis in single shell weight (+29.3%) with least filament denier (10.86 d), while the length of silk filament was moderate (1546 m) and the most important commercial trait, total silk yield was the least (35.0 g), specify its inadequacy for commercial sustenance. Though, the hybridization impact was positive with mid parent heterosis in shell weight of Daba×Raily the moderate filament denier, the least filament length and total silk yields show its lowest overall performance among the evaluated F_1 hybrids.

However, the better performance of parents during commercial crop season than the parents of seed crop and superior performance of F_1 hybrids over the parents of both crops, in general indicate the hybridization influence on silk productivity and quality. These observations are corroborating with the findings earlier reported by Moorthy *et al.* (2007), Doddaswamy *et al.* (2009) and Reddy *et al.* (2009c). Further, the assessment and comparison made among the parents of both crop seasons and the F_1 hybrids of commercial crop season has nullified the role of environment on the performance of parents and F_1 hybrids and to arrive at the actual influence of hybridization and relative heterosis on the silk productivity and quality.

CONCLUSION

The superior performance of F_1 hybrids in single shell weight, total silk yield and length and denier of silk filament indicates the influence of hybridization as positive heterosis and the study infers the commercial advantage and application feasibility of Daba×Jata and Jata×Daba F_1 hybrid combinations during commercial crop rearing season for optimal tasar silk productivity with silk filament denier at least better than one of the parents in F_1 hybrid combination.

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