New Research Project Proposal
on

DEVELOPMENT OF THERMOTOLERANT BIVOLTINE BREEDS / HYBRIDS OF SILKWORM, *BOMBYX MORI* THROUGH MARKER ASSISTED SELECTION

By

Principal Investigator: **N. Chandrakanth, Scientist -B**
Co-Investigator : **Dr. A. K. Verma, Scientist-D**
Shri. N. B. Kar, Scientist-D

Central Sericultural Research & Training Institute
Central Silk Board: Ministry of Textiles: Govt. of India
Berhampore – 742 101, Murshidabad, West Bengal, INDIA

Budget: 12.6 Lakhs

2016-17

PROFORMA-I

PART-1: GENERAL INFORMATION

1	Name of the Institute/ University/Organization submitting the project proposal	Central Sericultural Research & Training Institute, Central Silk Board, Ministry of Textiles, (Govt. of India), Berhampore - 742 101, Murshidabad - Dist., West Bengal.
2	Status of the Institute	Research and training institute under Central Silk Board, Ministry of Textiles, Govt. of India
3	Name and designation of the executive authority of the Institute/University forwarding the application	Dr.KanikaTrivedy, Director, CSR&TI, Central Silk Board, Berhampore - 742 101, Murshidabad - Dist., West Bengal.
4	Project Title	**"Development of thermotolerant bivoltine breeds / hybrids of silkworm, B*ombyx mori* through marker assisted selection"**
5	Category of the project	R & D
6	Specific Area	Silkworm improvement
7	Duration	5years
8	Total Cost (Rs.)	Rs. 12.6Lakhs
9	Is the project Single Institutional or Multi-institutional	Single Institutional
10	If the project is multi-institutional, please furnish the following :	NA

11. (a) Project Summary:

Silkworm being a poikilothermic insect, its growth, development and ultimately silk yield is adversely affected by high temperature conditions prevailing in tropical countries like India especially during summer. This effect is more pronounced in bivoltine breeds than multivoltine incurring significant loss to the bivoltine sericulture. In order to increase the bivoltine silk production in India, there is a need to develop thermotolerant bivoltine silkworm breeds / hybrids that can be reared throughout the year, including summer season. Over the years, conventional breeding efforts have led to the development of a large number of bivoltine silkworm hybrids, of which few have been under commercial exploitation. However, the genetic potential of crop production are constantly threatened by

environmental stresses *viz.*, biotic and abiotic factors that reduce crop yield and quality. Hence development of silkworm hybrids tolerant to abiotic stress is necessary to combat the climate hazards, low-input cum low management situation.

In this context, modern breeding techniques involving biotechnological aspects like marker assisted selection has been successfully applied in improving yield of several crops and live stocks. Hence, the situation warrants the need to reorient the breeding approaches using molecular biological tools on the available genetic stocks to supplement the conventional practices adopted in the breeding programs. Of late, genome analysis of mulberry silkworm using molecular markers has been initiated in India and elsewhere.

A total of 50 bivoltine germplasm is available at CSRTI, Berhampore. Out of which 10 bivoltine breeds has been screened for thermotolerance by rearing at 36 ± 1°C, in this study, the remaining 40 bivoltine germplasm will be screened for thermotolerance by rearing at 36 ± 1°C and 85 ± 5% relative humidity. The breeds with contrasting response to thermo-tolerance based on pupation rate will be selected and their F_1 and F_2 progeny will be developed. The DNA markers (Microsatellites) showing polymorphism between the selected parents will be used to screen their F_1 and F_2 progeny to determine the closely linked marker to thermotolerance. Thereafter the identified DNA marker linked to thermotolerance will be used for screening of breeds available in the germplasm stocks tolerant to high temperature. The breeds with high pupation rate at high temperature and presence of thermotolerant banding pattern will be selected as tolerant and donor parent while the breeds with susceptible banding pattern and high productivity will be selected as productive and recurrent parent. The tolerant and productive bivoltine breeds will be crossed followed by backcrossing and Inbreeding. In each generation progeny would be selected based on DNA markers linked to thermotolerance. The developed breeds and hybrids will be evaluated at high temperature (36 ± 1°C) and humidity conditions (85 ± 5%) and will be short-listed based on temperature tolerance and post cocoon parameters.

12. PART II: PARTICULARS OF INVESTIGATORS

12a)	Name:	Shri. N. Chandrakanth
	Date of birth	24-04-1986
	Sex (M/F)	M
	Indicate whether Principal Investigator/Co-investigator	Principal Investigator

	Designation	Scientist- B
	Department	Silkworm Breeding and Genetics Laboratory
	Institute/University and address	Central Sericultural Research and Training Institute, Central Silk Board, Ministry of Textiles, Govt. of India, Berhampore-742 101
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12b	Name:	Dr. A. K. Verma
	Date of birth	28.12.1960
	Sex (M/F)	M
	Indicate whether Principal Investigator/Co-investigator	Co-investigator
	Designation	Scientist-D
	Department	Silkworm Breeding and Genetics Laboratory
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12c	Name:	Shri. N. B. Kar
	Date of birth	1959
	Sex (M/F)	M
	Indicate whether Principal Investigator/Co-investigator	Co-investigator
	Designation	Scientist-D
	Department	Reeling section
	Institute /University and address	Central Sericultural Research and Training Institute, Central Silk Board, Ministry of Textiles, Govt. of

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13.	No of projects being handled by each investigator at present	
	Shri. N. Chandrakanth	01
	Dr. A. K. Verma	06
	Shri. N. B. Kar	04
14.	Proposed Research Fellows	-

PART III: TECHNICAL DETAILS OF THE PROJECT

15. Introduction

Sericulture as a viable proposition, which can be realized through many major factors coupled with exploitation of superior breeds to the optimum potential (Nirmal Kumar *et al*., 1999). Over the years, Sericulture research in India has resulted in increase of silk production, but silk quality remains low due to inherent defects of multi x bi cocoons, from which 90% of the silk is produced (Datta, 1984). To overcome the problem in production of quality silk and also to produce silk of International grade, there is necessary to rear bivoltine hybrids in India. Being a tropical country, bivoltine cannot be reared throughout year due to variable climatic conditions. In recent years, productive and superior quality silk producing bivoltine hybrids have been evolved for commercial exploitation (Basavaraja*et al.,* 1995; Datta*et al.*,2000a, b; 2001). But these hybrids have been recommended for rearing during favourable seasons of the year (Aug-Feb) in some parts of India. Hence, silkworm breeds/ hybrids tolerant to fluctuating tropical climatic conditions for rearing throughout year are necessary (Naseema Begum *et al.,* 2002).

Recent advances in the field of molecular genetics have contributed a number of DNA markers in plant as well as insect research with varying properties and usage. Marker assisted selection (MAS) is emerging as a very promising strategy for increasing selection gain (Knapp, 1998).If sufficient mapping information is available, MAS can dramatically shorten time required for genetic improvement in any crop and silkworms as well. Moreover, if a closely linked marker has been found, it is possible to screen large numbers of samples for rapid identification of progeny that carry desirable characteristics (Zhu *et al*., 2003). In this

project attempt will be taken to develop bivoltine silkworm breeds / hybrids tolerant to high temperature for utilizing them in fluctuating environment conditions of tropics without compromising the silk quality using marker assisted breeding.

15.1. Definition of the problem

In India, in order to increase bivoltine quality silk, previous studies conducted to develop thermotolerant bivoltine breeds / hybrids included conventional breeding based on phenotypic information that has not yielded the anticipated results. Therefore, the biotechnological tools must be integrated to develop thermotolerant silkworm breeds / hybrids. One of the main approach of breeding in molecular biology is marker assisted breeding. This approach includes the identification of DNA markers linked to thermo-tolerance paving the way for their application in silkworm breeding to develop hardy bivoltine silkworm breeds / hybrids suitable for rearing throughout the year including summer season.

15.2. Origin of the proposal /Rationale of the study

West Bengal is one among the traditional states practicing sericulture in India. West Bengal experiences extreme variations in temperature, relative humidity and rainfall. Therefore, popularization of bivoltine silkworm breeds in Eastern India, especially in West Bengal is difficult owing to the prevalence of adverse environmental conditions. In Eastern India, the multi x bi hybrids are very much popular in the autumn (Nov-Dec) and spring (Feb-Mar) crop seasons. However, during autumn (Nov-Dec), the major commercial crop season of this region especially in the major silk producing state West Bengal, even the required quantity of multi x bi hybrid layings production is not possible, mainly due to unavailability of bivoltine male seed cocoon. Since, the preceding seed crop (Sept-Oct) experiences high temperature, high humidity and unpredictable rainfall, which threat rearing of bivoltine parent silkworm (Moorthy *et al*., 2007). Keeping this in view, some bivoltine breeds / foundation crosses *viz*., SK6 x SK7, B.Con1 x B.Con4 were evolved through conventional breeding. Presently, foundation crosses SK6 x SK7 and its reciprocal are been preferred to be reared at even unfavourable seasons. Though these foundation crosses fairing well in adverse condition, it lacks productivity. Therefore, it is a need of the hour to develop bivoltine breeds which can be reared unfavourable season also has productivity.

Despite the continued yield improvement from conventional breeding, new biotechnological approach such as Marker assisted Selection (MAS) gained momentum in

plant and animal breeding programme. But success of MAS depends on availability of DNA marker for trait of interest. Recently, Chandrakanth *et al*., (2015) identified five microsatellite markers linked to thermotolerance in silkworms. Using this markers donor and recurrent parents were selected and breeding initiated to develop thermotolerant breeds employing MAS. The identified markers were found only on 8^{th} chromosome. Since, thermotolerance is polygenic, some marker may be present on other chromosome too. Therefore, in this study attempt will be made to identify some more markers for thermotolerance. After identification, the markers would be used for selecting parents followed by marker assisted selection in breeding programme. Hence in this project attempt will be made to identify additional DNA markers linked to thermotolerance and use them in marker assisted selection for developing thermotolerant bivoltine breeds/ hybrids that can be reared in fluctuating climatic conditions of West Bengal.

15.3. Relevance to the current issues

Sericulture in India is predominantly practiced in hot tropical regions; rearing of silkworms under such conditions will adversely affect pre- and post-cocoon parameters. Therefore, F_1 hybrids developed by crossing females of native multivoltine and males of exotic bivoltine breeds as parents are popular, because these cross breeds are more tolerant to high temperature but produce non-gradable silk (Lakshmi *et al*., 2011). Hence, in order to increase the production of gradable silk with superior quality in India, there is a necessity for the development of thermotolerant bivoltine silkworm breed/hybrid, which can be reared throughout year including summer season in India. However, it is a difficult task because the thermotolerance trait in silkwormis influenced by genetic and environmental factors. The earlier attempts made by many silkworm breeders have led to the development of new bivoltine breeds and hybrids. Though, they performed well under controlled laboratory conditions but failed to sustain at the farmers' level (Suresh Kumar *et al*., 2011). One of the possible reasons for the failure of popularization of these breeds was the selection of parents was solely based on their phenotypic performance without considering the genetic factors. However, parent selection based on per se performance, and genetic marker may be better option to obtain a wide genetic base, which can yield superior segregants enabling effective selection during the course of evolution of lines suitable for tropical regions (Moorthy *et al*., 2007). Therefore, in this project attempt will be made to identify the DNA markers (SSR / ISSR) linked to thermotolerance that can be used in developing thermotolerant bivoltine breeds/ hybrids which can withstand fluctuating climatic conditions of West Bengal.

15.4. Expected outcome

1. Hardy bivoltine silkworm hybrids suitable for rearing throughout the year including summer season of the tropics will be developed.

2. DNA markers (SSR / ISSR)linked to thermotolerancewill be identified and will be applied in the breeding program.

15.5. Objectives

1. Identification of DNA markers (SSR / ISSR) linked to thermotolerance.

2. Development of thermotolerant bivoltine silkworm breeds / hybrids through DNA marker assisted selection and their evaluation.

16. Review of current status of research and development in the subject

16.1.International Status

With the advancement in the genomics and availability of new molecular tools, silkworm genome has been greatly explored by successively constructing molecular linkage maps using markers like random amplification of polymorphic DNAs (RAPDs), amplified fragment length polymorphisms (AFLPs), restriction fragment length polymorphisms (RFLPs), microsatellites and single nucleotide polymorphisms (SNPs) (Shi *et al*., 1995; Promboon, *et al*., 1995; Yasukochi, 1998; Tan *et al*., 2001; Miao *et al*., 2005; Yamamoto *et al*., 2006), laying a strong foundation for mapping *B. mori* genes associated with the traits of commercial importance. Among these markers, microsatellites, also known as simple sequence repeats (SSRs) have gained considerable interest because of their reproducibility, multiallelic nature, codominant inheritance and good genome coverage (Powell, 1996). SSRs are tandem repeats of 1-6 bp of DNA sequence originated primarily due to slipped strand mispairing (Levinson and Gutman, 1987) and subsequent errors during DNA replication/ repair/ recombination (Katti *et al*., 2001) or unequal crossing over between sister chromatids (Innan *et al*., 1997).

Among insects, *Drosophila* has been used extensively for understanding the physiological and genetic basis of thermal tolerance variation in ectotherms (Huey and Kingsolver, 1993; Krebs and Loeschcke, 1995). The heat resistance ability have been greatly correlated with the variations in the *hsr-omega*, *hsp70* and *hsp68*, all of which map to the right arm of chromosome 3 (McColl *et al*, 1996). Furthermore, Rashkovetsky *et al* (2006) found difference in thermotolerance between flies from the two slopes of the canyon is

ongoing and robust and concluded that the inducible *Hsp70* is responsible for a substantial portion of inducible thermotolerance between the flies of two slopes of the canyon.

With regard to mulberry silkworm, *Bombyx mori*, more than 400 visible mutations have been placed in the linkage map (Doira, 1992), which represent 217 loci consisting of mostly morphological and few enzyme markers. The number of loci mapped so far is insufficient for a thorough understanding of the genome and for the analysis of the quantitative trait loci (QTLs) for important commercial characters in silkworm. Hence, the development of DNA based genetic markers was initiated in the 90s and preliminary linkage map using RFLPs (Shi *et al.,* 1995) and RAPD map of 169 loci (Promboon *et al.,* 1995) have been constructed. Polymorphism and analysis of Prothoracitrophic hormone (Shimada *et al.,* 1994) and genes of diapause hormone (Pinyarat *et al.,* 1995) have been carried out by PCR based primers. Recently a dense genetic map of silkworm covering all chromosomes based on 1018 RAPD markers has been published (Yasukochi, 1998). Further, a RAPD marker linked to densonucleosis refractoriness gene 'nsd-1' & 'nsd-2' has also been identified (Abe *et al.,* 1995, 1998, 2000). Of late, an AFLP map of the silkworm has been constructed with 356 markers (Tan *et al.,* 2001).Phenotype variations in a particular trait are likely to be linked to the microsatellite polymorphism present in the *B. mori* genome. For instance, Li *et al.* (2006) mapped the densonucleosis non-susceptible gene (*nsd-Z*) on 15th chromosome of *B. mori* by using SSR markers. Further, Zhao *et al.* (2006) mapped BmNPV-resistance gene of *B. mori* and also 3 SSR markers linked to I (yellow blood inhibitor) gene was identified on 9th chromosome of *B. mori* (Li *et al.*, 2008).

So far little information is available on the molecular marker based investigation for the analysis of QTL controlling yield traits or disease resistance in silkworm other than nsd-1. Lu *et al.* (2004) employed AFLP technique for mapping the QTLs for the cocoon weight, cocoon shell weight, ratio of cocoon shell, weight of pupae and 11 QTLs were detected based on the constructed linkage map.Recently, the 6× and 3× draft genome sequences generated respectively by Chinese and Japanese group were merged (Xia *et al.*, 2004; Mita *et al.*, 2004). This has resulted in a new silkworm genome assembly size of 432Mb with estimated genes of 14,623. The information on silkworm genomic DNA sequences is deposited as a SilkDB (http://www.silkdb.org) which is publicly accessible. Additionally, the annotated protein sequences are also deposited in SilkDB. Totally 11,104 of full-length cDNAs and 4, 08,172 expressed sequence tags (ESTs) have been deposited publicly in KAIKObase database (http://sgp.dna.affrc.go.jp/KAIKObase/) of Silkworm Genome Database. There is a SilkSat

database designed and maintained at Centre for DNA Fingerprinting and Diagnosis (CDFD), Hyderabad exclusively for using silkworm microsatellite sequences and analysis. Sericulturally advanced countries like in China, where climatic conditions similar to tropical climate were able to develop season specific (Summer & Autumn) bivoltine silkworm breeds / hybrids following the technique of hybridization, rearing at high temperature & humidity and QTL selection(Shao *et al.,* 1987; Shao, 1989; He *et al.,* 1991; Chen *et al*., 1994; Zhang *et al*., 1994).

16.2.National Status

During JICA period efforts were made by CSRTI, Mysore for the development of temperature tolerant bivoltine breeds has led to the development of robust bivoltine hybrids like CSR18 x CSR19, 5HT x 8HT and SR1 x SR5 for rearing in high temperature conditions of summer (Suresh Kumar *et al.,* 2002; Sudhakara Rao *et al*., 2006). Even though they are known for their productive merit, absence of genetic plasticity to buffer against the tropical environmental stresses acts as a constraint to tap the full economic potential of these hybrids. Earlier work has clearly shown the prospects of using digestive amylase as a marker in silkworm breeding due to its wide genetic diversity, role in better digestibility and high survival (Datta and Ashwath, 2000). A breeding scheme employing the new strategy of marker assisted selection was designed and amylase enzyme of Pure Mysore and Nistari were introgressed into CSR2 and CSR5 races respectively. The study has resulted in the evolution of the robust hybrid GEN3 x GEN2 (Ashwath *et al.,* 2001; 2002) and the field testing of over 30,000 dfls of the hybrid has recorded an average yield of 63kg / 100 dfls.

Silkworm genome has been explored greatly by constructing molecular linkage maps using markers like RAPDs, AFLPs, RFLPs, SSRs (Microsatellite) and SNPs (Single nucleotide polymorphisms) (Shi *et al*., 1995; Promboon *et al*., 1995; Yasukochi, 1998; Tan *et al*., 2001; Miao *et al*., 2005; Yamamoto *et al*., 2006), thereby establishing a strong base for mapping genes associated with the commercial traits of *B. mori* (Zhao *et al*., 2010). The PCR based RAPD and also DNA fingerprinting with minisatellite probes have been taken up to study the DNA profiling of silkworm genotypes (Nagaraju and Nagaraju, 1995; Nagaraju*et al.,* 1995). Genetic characterization by simple sequence repeats (SSR), inter-SSR (ISSR) has been taken up in 13 silkworm strains (Reddy *et al.,* 1999a, 1999b; Nagaraju *et al.,* 2001). These studies have revealed clearly separated the bivoltine and polyvoltine strains with specific alleles. These results have indicated their potential as generate markers that are linked to traits of interest in silkworm.

Considerable amount of studies also have been done in respect of thermotolerance in silkworm, *Bombyx mori* and high temperature effect on commercial traits (Joy and Gopinathan, 1995; Vasudha *et al.,* 2006; Moorthy *et al.,* 2007). Findings has revealed that thermotolerancevaries with strains, instars and tissues with respect to expression of heat shock protein. **Chandrakanth and Manthira Moorthy *et al.*** (2015) studied the expression of nine *Hsps* in thermo susceptible, thermotolerant bivoltine and thermotolerant polyvoltine silkworm breeds after heat shock and recovery period and confirmed the role of HSP expression in thermotolerance of silkworm. Srivastav *et al.* (2007) reported that out of ISSR markers showing correlation with pupation rate after thermal treatment, marker 808_{3000} showed highest and significant correlation with pupation rate among the silkworm races. Recently, **Chandrakanth *et al.*** (2015) identified five microsatellite markers linked to thermotolerance on 8^{th} chromosome of silkworm. From the above literature survey, it is found that a systematic study is required to identify a DNA marker in relation to thermotolerance which can be used as a tool in marker assisted breeding.

16.3. Importance of the proposed project in the context of current status

Rearing bivoltine silkworm breeds during summer season results in significant loss to the farmers as bivoltine breeds. Therefore, introduction of thermotolerant bivoltine breeds, which can produce quality silk is the need of the hour in tropics. This can be achieved by breeding bivoltine silkworm breed through marker-assisted selection which can significantly enhance genetic gain for traits where the phenotypic information is difficult to evaluate because of its dependence on specific environmental conditions. Molecular markers can be used to increase the probability of identifying truly superior genotypes by focusing testing resources on genotypes with the greatest potential (i.e. early elimination of inferior genotypes), by decreasing the number of progeny that must be screened to recovery of a given level of gain and by enabling simultaneous improvement for traits that are negatively correlated. Considering the problem of prevailing environmental conditions during summer months and advantages of using MAS in breeding, it is possible to develop thermotolerant bivoltine breeds / hybrids by utilizing the biotechnological tools that can be reared throughout the year including summer seasons of West Bengal.

16.4. Anticipated products & Processes /technology packages information or other outcome from the project and their expected utility

Hardy bivoltine silkworm breeds / hybrids which can be reared throughout the year even during summer season.

16.5. Expertise available with the proposed investigating group/institution in the subject of the project

The investigating group has knowledge and experience in breeding thermotolerant silkworm hybrids as well as in DNA marker based molecular biological work.

16.6 List of 5 experts in India in the proposed subject area

Sl.No.	Name	Designation	Address
1	Dr. H. K. Basavaraju	Scientist-E (Retired)	APSSRDI, Hindupur
2	Dr. Salil Kumar Das	Scientist-E (Retired)	CSR&TI, Berhampore, West Bengal
3	Dr. G. S. Rajanna	Scientist-D	KSSRDI, Thalaghattapura, Bengaluru, Karnataka
4	Dr.G. Subramanya	Professor	Dept. of Sericulture, University of Mysore, Mysuru, Karnataka
5	Dr. R.N. Chatterjee	Professor	University of Calcutta, Kolkata, West Bengal

17. Work plan

17.1. Methodology

Phenotypic screening of 40 unscreened bivoltine breeds at CSRTI, Berhamporeby rearing at 36 ± 1°C and 85 ± 5% relative humidity

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Molecular screening of 40 unscreened bivoltine breeds by existing microsatellite markers linked to thermotolerance

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Based on the pupation % and tolerant banding pattern thermo-tolerant and -susceptible breeds will be selected

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DNAs of thermo-tolerant and -susceptible breeds will be screened for polymorphism using SSR / ISSR primersby PCR

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Development of F_1 and F_2 generation by crossing between identified tolerant and susceptible breeds

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Polymorphic SSR / ISSRmarkers linked to the thermotolerance traitwill be identified by Bulked segregant analysis between F_2bulks

↓

Suitable parents will be selected by phenotypic and molecular screening and breeding of thermotolerant bivoltine lines through marker assistedbackcrossing (Appendix 1)

↓

Development and evaluation of thermotolerant hybrids at 36± 1°C

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Based on pupation %(65% pupation) and post cocoon parameters at 36 ± 1°Cshort listing of the hybrids will be done.

17.2. Organization of work elements

Name of the Scientists	Designation	Time	Organization of work elements
N. Chandrakanth	Scientist-B	60%	Planning, execution and monitoring of the project
Dr. A. K. Verma	Scientist-D	20%	Assisting in planning, execution and monitoring of the project
N. B. Kar	Scientist-D	20%	Assessment of post-cocoon parameters

17.3. Proprietary/patented items, if any expected to be used for this project

Silkworm breeds / hybrids

17.4. Suggested plan of action for utilization of research outcome expected from the project

The developed silkworm breeds / hybrids tolerant to high temperature environments will be recommended for rearing throughout the year even during summer season.

17.5 Time schedule of activities giving milestones

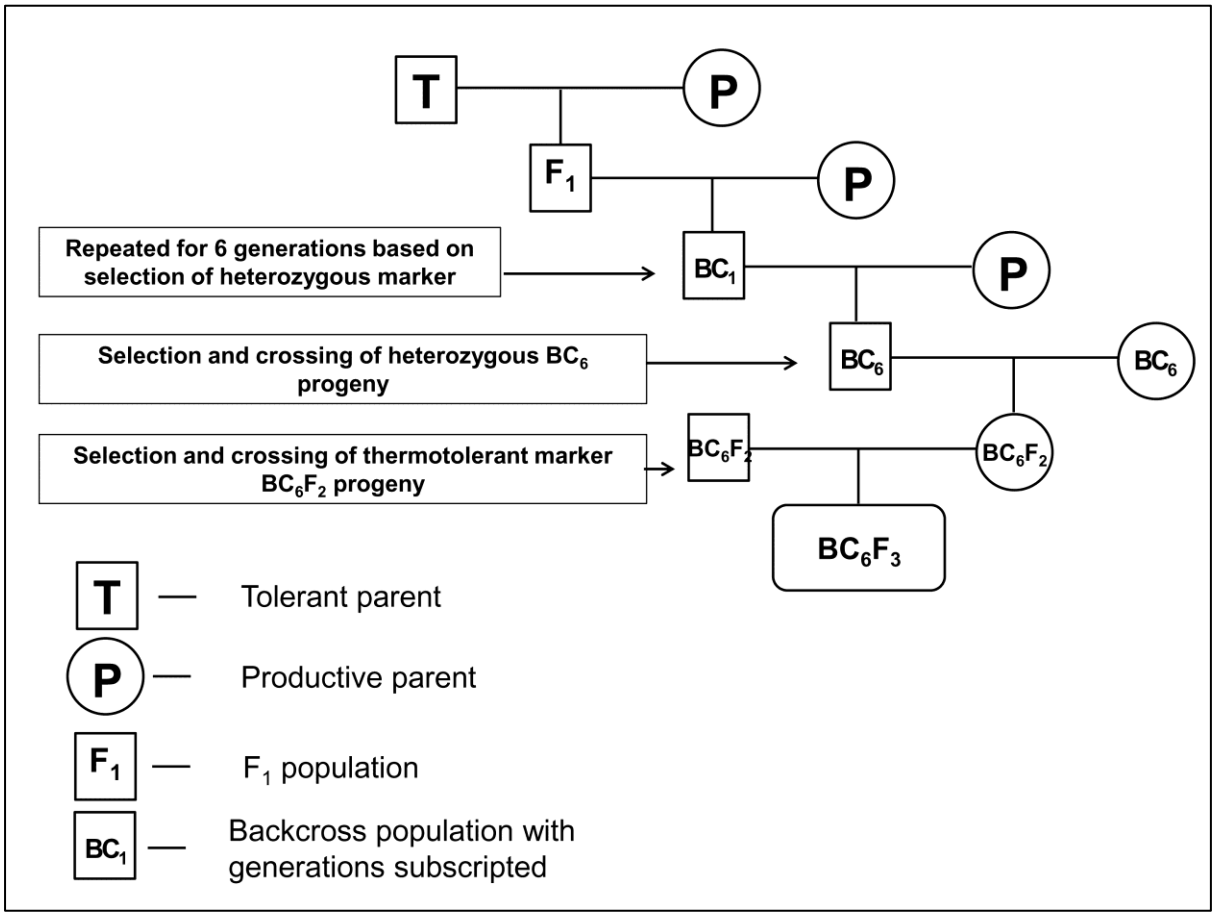
Sl No	Name of the milestone	Expected (Month/Year)	
		Start	Completion
1	Selection of parents and screening of parents DNA having contrasting characterswith respect to thermotoleranceby DNA markers (SSR / ISSR) for identification of informative markers.	May- 2016	April- 2017
2	Screening of informative markers throughbulked segregant analysis in the F_2 bulks in comparison with parents and F_1s to identify markers linked to the thermotolerance trait.	May- 2017	April- 2018
3	Selection of parents and breeding process employing DNA marker assisted selection.	May- 2018	April- 2020
4	Development and evaluation of thermotolerant hybrids.	May- 2020	Dec-2020

5	Compilation of results and submission of final report	Jan-2021	April- 2021

18.6. Project implementing agency/agencies

Name of the agency	Address of agency	Proposed research aspects	Proposed amount (Lakhs)	Cost sharing %
Central Silk Board	CSRTI, Berhampore	--	12.60	Nil

Appendix- 1



PART IV: BUDGET PARTICULARS

19.BUDGET (in Rupees): [In case of multi –institutional projects, the budget details should be provided separately for each of the institute]

A. Non-Recurring (e.g. equipments, accessories, etc.)

(In Lakh rupees)

S.No.	**ITEM**	1st Year	2nd Year	3rd Year	4th Year	5th year	Total
1.	Themocycler (PCR engine)	3.50	-			-	3.50
2.	Vertical Gel Electrophoresis unit	0.65	-			-	0.65
3.	pH meter	0.40	-			-	0.40
	Sub-total A	**4.55**	-			-	**4.55**

B. Recurring: B1. Manpower: **(In Lakh rupees)**

S.No.	Position	No.	Consolidated emoluments	1st Year	2nd Year	3rd Year	4th Year	5th Year	Total
1.	JRF								
-	HRA								
-	Medical								
	Sub-total B1								

B2. Consumables **(In Lakh rupees)**

Sl.No.	ITEM	1st Year	2nd Year	3rd Year	4th Year	5th year	Total
1.	Chemicals	2.00	1.50	1.50	1.00	-	6.00
2.	Rearing material & Stationary	0.20	0.20	0.30	0.30	0.40	1.40
	Sub-total B2	**2.20**	**1.70**	**1.80**	**1.30**	**0.40**	**7.40**

Other Items:

Sl.No.	**ITEM**	1st Year	2nd Year	3rd Year	4th Year	5th Year	Total
B3	Travel	-	-	0.10	0.10	0.05	0.25
B4	Contingency	0.10	0.10	0.10	0.05	0.05	0 .40
B5	Overhead Charge	-	-	-			-
	Sub-Total	**0.10**	**0.10**	**0.20**	**0.15**	**0.10**	**0.65**
	Sub-total (B1+B2+B3+B4+B5+etc.)	**0.00**	**7.40**	**0.25**	**0.40**	**0.00**	**8.05**

Budget: **4.55 (Non-recurring) + 8.05 (Recurring): .12.60 lakhs**

PART V: EXISTING FACILITIES

19. Available equipment and accessories to be utilized under the project

Sl.No.	Name of equipment/accessory	Make	Model	Funding agency	Year of procurement
1	Gel documentation system	UVP		CSB	1997
2	Refrigerated water batch circulator	Jeica Tech	RW 1025 G	CSB	2004
3	Ice flakers	Simag	SPR 80	CSIR	2005
4	pH meter	Thermo	420 A	CSB	2005
5	Deep Freeze (-80°C)	Hareaus	HFU 486	CSIR	2006
6	Vertical gel electrophoresis system	Omega, Japan	-	CSB	2002
7	UV spectrophotometer	Shimadzu	-	CSB	1994
8	Micro-pipettes	Eppendorf and Tarson	-	CSB	2004
9	Micro centrifuge	Hermile	-	CSB	2002
10	Refrigerated Centrifuge (High speed)	Sorvall	-	CSB	1992
11	Refrigerators	LG	GLD 32	CSB	2005
12	Computer	HCL	-	CSB	2004
13	Electronic top loading balance	Sartorious	GE 812	CSIR	2005
14	Deep freeze (-35°C)	Remi	RQFV 265 (D)	CSB	2006
15	Environmental chamber	SD Instruments	HTC 3005	CSB	2014

PART VI: REFERENCES

Abe, H., T. Shimada., T. Tsuji., T. Yokoyama., T. Oshiki and M. Kobayashi (1995) Identification of random amplified polymorphic DNA linked to the densonucleosis virus type-1 susceptibility gene of the silkworm, *Bombyx mori. J. Seric. Sci. Jpn.*, 64:262-264 (In Japanese).

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Basavaraja, H.K., S. Nirmal Kumar, N. Suresh Kumar, N. Malreddy, KshamaGiridhar, M.M. Ahsan and R.K. Datta (1995) New productive bivoltine hybrids. *Indian Silk*, 34(2): 5-9.

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PART VII: BIODATA OF PRINCIPAL INVESTIGATOR

1. Full Name (in Block Letters) : N CHANDRAKANTH
2. Designation : Scientist - B
3. Department /Institute /University : CSRTI, Berhampore
4. Address for communication : SBG, CSRTI, Berhampore
5. Date of birth : 24/04/1986
6. Sex : Male
7. Education onwards & (Post Graduation onwards & Professional Career)

Name of the university	Degree Passed	Year of Passing	Subjects taken with Specialization	Class/ Divn.
Punjab Technical University, Jalandhar	M. Sc.	2009	Biotechnology	I
University of Mysore, Mysore	Ph.D.	2016	Biotechnology (Submitted)	-

8. Awards: [Not required for house personnel]:

Year	Award	Agency	Purpose	Nature
Nil	Nil	Nil	Nil	Nil

9. Position Held / Research Experience in various institutions:
[Not required for in –house personnel]
10. Memberships/Fellowships: [Not required for in-house personnel] :
11. Patents: [Not required for in-house personnel]:
12. Publications (Number only): 12
Books: 01
Research Papers, Reports: 11
General articles: Nil

13. Project(s) submitted / being pursued / carried out by Investigator:

Sl.No.	Title of the Project	Funding agency	Duration From and To	No of Scientists /Associates working under the project	Total approved cost of the project (Rs.in lakh)
Nil	Nil	Nil	Nil	Nil	Nil

14. Highlights of outcome / progress of the project (s) handled during the past 10 years their outcome and utilization (in 200 words).
NIL

15. List of publications

- **Chandrakanth N**, Moorthy SM, Ponnuvel KM and Sivaprasad V (2015) Identification of microsatellite markers linked to thermotolerance in silkworm by bulk segregant analysisand *in silico* mapping. *Genetika* 47(3): 1063-1078. (IF: 0.4)
- **Chandrakanth N**, Ponnuvel KM, Moorthy SM, Sasibhushan S and Sivaprasad V (2015) Transcript analysis of heat shock protein genes in Silkworm, *Bombyx mori* in response to heat shock. *Eur. J. Entol.*112(4): 676–687.(IF: 1.06)
- **Chandrakanth N**, Moorthy SM and Sivaprasad V (2015) Performances of F_2 and backcross populations in silkworm, *Bombyx mori*under high temperature. *Annals of Biological Research* 6(9):1-6.
- **Chandrakanth N**, Moorthy SM, Kariyappa, Ponnuvel KM and Sivaprasad V (2015) Reeling performances of F_2 and backcross populations under high temperature condition. *Journal of Entomology and Zoology Studies* 3(6): 219-222.
- **Chandrakanth N,** Moorthy SM, Anusha P, Dayananda, Ashwath SK, Kumar V and Bindroo BB (2014)Evaluation of genetic diversity in silkworm (*Bombyx mori L.*) strains using microsatellite markers.*IJBAF* 2(3): 73-83.
- **Chandrakanth N**, Anusha P and Moorthy SM (2014) Analysis of genetic variability in some silkworm strains of *Bombyx mori* L. through isoenzyme markers. *IJAPBC* 3(2).
- Moorthy SM and **ChandrakanthN** (2015) Analysis of phenotypic diversity and protein polymorphism in some silkworm breeds of *Bombyx mori* L. *Mun. Ent. Zool.* 10 (2): 478-485.
- Moorthy SM, **Chandrakanth N**, Rao ASK, Kumar V and Bindroo BB (2013) Genetic diversity analysis using RAPD marker in some silkworm breeds of *Bombyx mori* L. *Annals of Biological Research* 4 (12):82-88.
- Uma Reddy B, **Chandrakanth N**, InduPriya S, VenkataNagalakshmi R and Usha B (2009) Isolation and characterization of faecal coliforms in street vended fruit juices and its safety evaluations: A case study of Bellary city, India. *Internet J of Food Safety* 11: 35-43.
- Sumathy R, Rao ASK, **Chandrakanth N** and Gopalakrishnan VK (2014) *Insilico* identification of protein-protein interactions in Silkworm, *Bombyx mori*. *Bioinformation* 10(2): 56-62.

PART VII: BIODATA OF CO-INVESTIGATOR

1.	Full Name (in Block letters)	DR. ANIL KUMAR VERMA
2.	Designation	Scientist-D
3.	Department/Institute/University	Silkworm Breeding Section, Central Sericultural Research & Training Institute, Berhampore(WB)-742101
4.	Date of birth	28.12.1960.
5.	Sex	Male.

6. Education (Post Graduation onwards & Professional careers)

Name of the University	Degree passed	Year of passing	Subjects taken with specialization	Class / Division
1. University of Kalyani, Nadia, West Bengal.	M.Sc.	1983	Zoology, Spl.: Entomology.	I
2. Bidhan Chandra KrishiViswavidyalaya, West Bengal	Ph. D	1990	Title of the Thesis- Studies on **whitefly** as vector of plant viruses in West Bengal.	-

7. Awards: [Not required for in-house personnel] Not applicable

Year	Award	Agency	Purpose	Nature

8. Working experiences:

Sl. No	Name of Employer	Designation	Designation		Nature of duty
			From	To	
1	Central Silk Board	SRA	12.10.1990	31.05.1996	Applied research in muga& Extension
2	Do	SRA	01.06.1996	31.05.1998	Research & Extension in Mulberry silkworm
3	Do	SRA/SRO	01.06.1998	31.05.2006	Research on Silkworm Breeding, Genetics & Molecular Biology
4	Do	Sc.-C	01.06.2006	31.07.2013	Research & Extension in Mulberry silkworm
5	Do	Sc.-C/ Sc.-D	01.08.2013	Till date	Research on Silkworm Breeding & Genetics

9. Publications :

- Research Papers : 21 (International- 5; National: 16)
- Abstracts in seminars etc. : 06
- Book : 02
- Technical Manual/Brochure: 07
- Technical Reports : 35
- PGDS Dissertation : 06

LIST OF IMPORTANT PUBLICATIONS RELATED TO SERICULTURE:

- Chattopadhyay, G. K., Sengupta, A. K., **Verma, A. K.,**Sen, S. K. and Saratchandra, B. **(2001 c)** Esterase isozyme polymorphism, Specific and nonspecific esterase, syngenic line development and natural occurrence of a thermo-stable esterase in tropical silkworm *Bombyx mori* L., ***Insect Biochem. Mol. Biol. 31: 1191-1199.***
- Chattopadhyay, G. K., Sengupta, A. K., **Verma, A. K.,**Sen, S. K. and Saratchandra, B. **(2001a)** Utilization of congenic line in silkworm breeding. In: ***Perspectives in Cytology & Genetics.*** G.K. Manna and Roy, S.C (edt). ***10: 717-724.***
- **Verma, A. K,**Chattopadhyay, G. K.;Sengupta. M, Sengupta, A. K. Das, S. K. and RajeUrs, S. (2003) Expression of heterotic genetic interaction among multivoltine backcross / congenic line for higher shell weight of silkworm *Bombyx mori* L. ***Inter. J. of Indust. Ent. (IJIE), Korea.*** **7(1): 21-27.**
- Chattopadhyay, G. K., Sengupta, A. K., **Verma, A. K.,**Sen, S .K. and Saratchandra, B. **(2001b)** Transgression of shell weight - A multigenic trait, through development of congenic breed in tropical silkworm *Bombyx mori* L. ***Sericologia. 41(1): 33-42.***
- Chattopadhyay, G. K., **Verma, A. K.,**Sengupta, M., Sengupta, A. K., Das, S. K and RajeUrs, S. **(2004)**α-and β-amylase isozyme possessor native proteins in tropical silkworm *Bombyx mori* L. ***Int. J. Indust. Entomol. 8(2): 21-27.***
- Chattopadhyay, G. K.;**Verma, A. K**.; Das, S. K. and Sarkar, A.(2005a) Acid phosphatase isozyme possessor native proteins pattern in Congenic breeds of silkworm, *Bombyx mori* L. In: *12th All India Congress of Cytology and genetics.* ***Institute of Science, Mumbai.*** **C-38.**

- **Verma, A. K,**Chattopadhyay, G. K.; Sengupta, M.; Sengupta, A. K.; Das, S.K. and RajeUrs, S. (2005) Heterobeltiotic genetic interaction between congenic and syngenic breeds of silkworm *Bombyx mori* L. ***Inter. J. of Indust. Ent. (IJIE), Korea.*** **11(2): 119-124**

- Chattopadhyay, G. K;**Verma, A.K.,** Das, S. K. and Sarkar. A. (2005b) Esterase-a Biochemical marker for quantitative traits of silkworm, *Bombyx mori* L. In: ***National Symposium on Development Dynamics. Indian Society of Developmental Biologists*****(Nov.23rd-25th).**Department of Zoology.University of Kalyani,Kalyani-741235. West Bengal.

- **Verma, A.K**., Suresh Kumar, N, Saha, A.K. and Nirmal Kumar, S. (2016) Heterotic Genetic interaction among high shell weight and high shell percentage multivoltine lines of silkworm Bombyx mori, L. J. Exp. Zool. India Vol. 19, No. 1, pp. 249-255.
- Chattopadhyay, G.K., **Verma, A.K**., Saha, A.K. and Nirmal Kumar, S. (2015) Specific differences among isozyme possessor native proteins in haemolymph of tropical multivoltine, bivoltine and developed congenic breeds of silkworm (Bombyx mori, L.). Biochem Cell Arch. Vol. 15, No. 2, pp. 549-555.
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12. Project(s) submitted / being pursued / carried out by Investigator: **18**
Project under progress: As PI-4 & as CI-2

13. Highlights of outcome / progress of the project(s) handled during the past 10 years, their outcome and utilisation (in 200 words):

Project	Outcome	Utilisation
1. Utilization of Syngenic lines for improvement of shell weight and survival in silkworm, *B. mori*. L	Isozyme based seven multivoltine and one sex linked bivoltine syngenic lines, four high cocoon shell weight multivoltine congenic breeds & four high survival bivoltine congenic breeds developed. Biochemical marker for high shell weight and high survival identified	The hybrids of the developed congenic breeds are under Post Authorization Trial.
2. Introgression of higher shell weight, higher survival character/gene through the development of multivoltine and bivoltine cogenic breeds and identification of biochemical marker in silkworm, *B. mori*. L.	Isozyme based three syngenic lines of Nistari and one high survival bivoltine congenic breeds developed. Two biochemical marker identified similar to above project.	The hybrids of the developed breeds are under Post Authorization Trial.
3. Institute Village Linkage Programme (IVLP)	Mulberry leaf yield (MT/ha/year) increases from initial 8 to 10.47(30.86%). Average Mulberry holding (in acre) increases from initial 0.5 to 0.66(32.00 %.). Average rearing capacity (DFLs/farmer/crop) increases from initial 75 to 125 (66.66 %.). Cocoon yield/ 100 DFLs (kg.) increases from initial 28.99 to 36.62(26.31 %.)	Plantation of High Yielding Variety like S1635. Plant to plant & row to row spacing- 2 ft.X 2 ft. Use of Plant Growth Hormone like Morizyme-B. Use of Vermicompost. Use of Biofertilizers like Nitrofert and Phosphofert to reduce the application of chemical nitrogen and phosphorus. Use of promising hybrids in place of existing one. Use of Bleaching Powder for

		general disinfection and Labex as bed disinfection. Use of dichlorovos for control of Whitefly infestation.
4. Validation trial of the Ready Reckoner of sulphur fertilizer application for obtaing targeted yields of mulberry	Soil from individual farmers analized to determine the extent of sulphur, based on this recommendation for sulphur application has been worked out.	Farmers are utilising this recommendation for use of sulphur in their field

Introduced a **method for introgression of a trait controlled by multiple genes** for developing

Congenic Breed (Chattopadhyay*et al.*, 2001a, b, 2005).

Developed **four promising silkworm breeds** viz., M Con.1, M Con.4 (Multivoltine), B Con.1, B. Con.4 (Bivoltine)

Identified **five promising hybrid** combinations.

M Con.1 x M Con.4, N x M Con.4 (Multivoltine x Multivoltine)

M Con.1 x B Con.4, M Con.4 x B Con.4 (Multivoltine x Bivoltine)

B Con. 1 x B Con.4 (Bivoltine x Bivoltine)

Registration of breeds: Six (6) congenic breeds viz., V^3 CB5-Con.Ow, V^3 M6DPC-Con.C, V^2 D6p-Con.Ow, V^2 D6p- Conc., V^2 D6p-Con.F and one sex limited breed (JPN $^{+HS}$) was send for registration at CSGRC, Hosur.

Biochemical study / Markers: Established that **amylase** is one of the most important enzymes in tropical silkworm having **positive correlation with high survival.**

It has been identified **224kDa Protein as a biochemical marker at pH-8.5 for high survival.** Theapparent native protein in haemolymph is the possessor of α-Est s are exclusively present in multivoltine.

It has been Identified that **180 kDa protein as a biochemical marker for high cocoon shell weight(at pH-8.5).** Theapparent native protein in haemolymph is the possessor of α-Est s and exclusively present in bivoltine.

β -amylase presence in haemolymph and digestive of Silkworm, *Bombyx mori* L. and **Identified specific and non-specific esterases** using α- and β-naphthyl-acetate separately as non-specific substrates. The non-specific β-esterase-Est-3 in haemolymph is a **thermo-stable enzyme (80 ± 1°C), which has been considered as one of the molecular factor for thermo-tolerance.**

Specific **Isozyme possessor native proteins** are associated with **non-hibernation and hibernation character** of silkworm has been identified some (CSIR Final report)

PART VII: BIODATA OF CO-INVESTIGATOR

1. NAME : Shri. N.B KAR
2. EMPLOYEE NO. : 003322
3. OFFICIAL DESIGNATION : Scientist-D (Reeling & Spinning)
4. PROJECT DESIGNATION : Co-Investigator
5. EXPERTISE AREA : Mechanical Processing of Textiles
6. INSTITUTE NAME : Central Sericultural Research & Training Institute, Berhampore-742 101, West Bengal.
7. INSTITUTE ADDRESS : Central Sericultural Research & Training Institute, Berhampora-742101, West Bengal.
8. TELEPHONE : 03482 – 251046, 251233, 253962
9. TELEX/E-MAIL : karnb@rediffmail.com
10. FAX : 03482 –251046
11. BIRTH YEAR : 1959
12. SEX : Male
13. EDUCATIONAL :

HIGHEST DEGREE (DEGREE ON WARDS	YEAR	UNIVERSITY	COUNTRY	SUBJECT
B. Sc. (Tech) in Textile Technology	1980	Calcutta	India	Spinning, Weaving, Dyeing & Printing, Fibre Science, Testing etc.
M.Sc(Tech) in Mechanical Processing of Textiles	1999	Calcutta	India	Spinning, Weaving, Fibre Science etc.

14. TRAINING UNDERGONE:

COURSE NAME/ SUBJECT	ADDRESS OF INSTITUTE	DURATION	PERIOD		SPONSORED AGENCY
			FROM	TO	
1. Refresher Training on Mulberry	CTR&TI, Ranchi	Three Weeks	1987		**CSB**
2.Tribal Orientation Training Programme	Tribal Research Institute, Ranchi	One Week	1987		
3.Trainers' Training Programme	CSTRI, Bangalore	Ten days	1992		
4.Entrepreneur Development Programme	NISIET, Hyderabad	Two weeks	1993		
5.Computer Training	CSR&TI, Berhampore	Two weeks	1994		
6)Orientation Training	CSTRI, Bangalore	One day	2001		

15. EXPERIENCE:

ORGANISATION/ INSTITUTE	DESIGNATION	DURATION	PERIOD		SUBJECT AREA	SIGNIFICANT ACHIEVEMENT
			From	To		
CTR&TI, Ranchi	SRA	4 yrs	1986 (July)	1990 (May)	Research on Tasar Reeling, Spinning & fiber technology	• Design & development of Reeling Cum Twisting machine (1987 – 1990). • Design & development of Studies on a User-Friendly 2-Spindle horizontal Tasar Reeling machine (1988 – 1990). • Studies on the weaving of Tasar raw, dyed & waste spun yarn for product diversification (1988 – 1990).
RSDTC, Fakirpur, Orissa	SRA	1¼ year	1990 (May)	1991 (Aug)	Tasar Research Extension & Training	Associated with Research extension under CTR&TI, Ranchi.
DCTC, Dabok, Rajasthan	SRO	App. 2½ years	1991 (Sept)	1994 (Mar)	Mulberry Research Extension & Training	• Associated with extension activities under NSP • Associated with Research extension under CSTRI, Bangalore
CSR&TI, Berhampore	SRO	3½ years	1994 (Mar)	1997 (Dec)	Research on Mulberry Reeling, Spinning &fibre Technology	• Designing of Reeling machine suitable for West Bengal condition to reel N, Nx G & N x Bi cocoons **(1988 – 1994)** Development of cocoon drying chamber suitable for West Bengal Condition **(1989 – 1994).** • Development of Water Correction Kit for cooking and reeling with reference to the states of eastern region **(1987 – 1994).**
Study Leave	SRO	2 years	1998 (Jan)	1999 (Dec)	Mechanical Processing of Textiles	Obtained M.Sc (Tech) degree from Calcutta University
DCTSC, Madhughat, Malda, West Bengal.	SRO	4¼ years	2000 (Jan)	2004 (Apr)	Mulberry Research Extension &	Associated with Research extension under CSTRI, Bangalore.

					Training	
CSR&TI, Berhampore	SRO	App. 4 years	2004 (May)	2006 (August)	Research on Mulberry Reeling, Spinning &fibre Technology	• Associated with 4 no. Ongoing & 5 no. Concluded Research Projects as CI. • Associated with 2 no. Ongoing & 4 no. Concluded Research Projects as non-CI. • Associated with 03 no. Ongoing & 4 no. Concluded Research Programmes.
CSR&TI, Berhampore	Scientist - C	App. 8 years	2006 (Aug)	2014 (Feb)	Research on Mulberry Reeling, Spinning &fibre Technology	
CSR&TI, Berhampore	Scientist – D	App. 2 years	2014 (Feb)	Till date	Research on Mulberry Reeling, Spinning &fibre Technology	

16. ACTIVCITY

a) PRESENT DISCIPLINE OF WORK **: Reeling & spinning**

b) PRESENT AREA OF WORK **: Post Cocoon Technology**

17. TIME ALLOCATION [IN %] & COST **:**

[NOTE:One week = 2%] [For one year]

SL. NO.	NATURE OF WORK	TIME %	NO. OF WEEKS	COST [RS. IN LAKHS]	COST [RS. IN LAKHS]

A] PROJECTS UNDER PROGRESS

SL. NO.	PROJECT CODE	PROJECT TITLE	RESEARCH TIME SPENT [%]	COST [RS.]
1	BAI(P) – 014	Studies on the Reelability of Multivoltine Hybrid Cocoons during adverse climatic condition in Eastern and Northeastern Region	50%	0.024
2	AIB - 3480	Development of Silkworm (Bombyx mori *L*) Breeds from a Gene Pool with Higher Genetic Plasticity	9%	0.024

3	AIB - 3466:	Development of Region Specific Bivoltine Breeds suitable for Highly Fluctuating Seasonally Variable Climatic Condition of Eastern & North-Eastern India	9%	0.024
4	Other Programmes		32%	0.024
5				0.172
		TOTAL	**100 ***	

B] COMPLETED PROJECTS

APR 3250: Development of Rearing Package for optimizing cocoon yield in West Bengal

APS 3238: Induction of Trimoulting in Bivoltine silkworm by physiologically active chemicals and their utilization as male parents for multi x bi hybrid production.

AIG 002: Utilization of Syngenic Lines for Improvement of Shell Weight & Survivals in *Bombyx mori-L*

PPA 3249: Effect of Integrated Plant Nutrition Systems on Mulberry Crop Production & Protection

PPA 3223 Updation of Improved Package of Practices for newly Authorized Mulberry Varieties under Irrigated Condition

AIB 3237 Utilization of Polyvoltine Breeds for Improvement of Survivals in Bivoltine Silkworms *Bombyx Mori*-L.

AIB 3291 Evaluation of viable Sex Limited Bivoltine Breeds of *Bombyx Mori-L.*

PPA 3358 Organic Farming in Mulberry-An Approach for Improvement of Silk Industry.

PPA 3366Development of Integrated Package for Raising Chawki Leaves & Young Age Silkworm Rearing for Successful Cocoon Crops

Other Projects &Programmes:

*** =** 100%IS THE TOTAL RESEARCH TIME WHICH IS EQUIVALENT TO THE % OF THE TIME SPENT ON RESEARCH AS INDICATED UNDER 'RESEARCH' AGAINST POINT NO. 17.

19. Highlight of the outcome/ progress of the project (s) handled during the past 10 years, their outcome and utilization (in 200 words):

The findings of the various research projects from the different sections particularly Silkworm Breeding and Genetics Section and Silkworm Physiology and RTI Section are finally substantiated by the post-cocoon assessment. Satisfaction of a reeler has the ultimate role for acceptance of a technology developed either by SBG or RTI. Part contribution of a reeler acts behind the success of a breeder. Previously eastern part of India reared only Nistari and Nx Bi dfls in 3-5 seasons according to the meteorological area. Now after introduction of various high yielding silkworm breeds, both multi x multi and multi & bivoltine the productivity of the farmers per unit laying and the rearing capacity of the farmers have been increased which also enable them to fetch higher income. A suitable alternative to Bivoltine component NB4D2 has been found and as

such SK6 x SK7 (FC) is going to replace NB4D2 successfully that can be reared throughout the year irrespective of climatic vagaries. Extensive study on breed development has also pave the way for some other combinations of silkworm races viz.; M.Con.1 x M.Con.4, Nistari x M.Con.4, M.Con.1 x B. Con.4 and M.Con.4 x B.Con.4 to replace the existing low productive breeds / hybrids. I was actively associated as co-investigator with all the projects during last seven years formulated by SBG or RTI section. Some work on formulation of an ideal package of practices of rearing during different climatic conditions has also come out with definite recommendation. Large scale testing of breed / hybrid at farm level, Cluster Promotion Programme at different zones has supported the breeds to become popular at commercial level.

PART VI: DECLARATION / CERTIFICATION

It is certified that

a. The research work proposed in the project does not in any way duplicate the work already done or being carried out elsewhere on the subject.

b. The same project has not been submitted to any other agencies for financial support.

c. The emoluments for the manpower proposed are those admissible to persons of corresponding status employed in the institute / university or as per the Ministry of science & technology guidelines (Annexure – III).

d. Necessary provision for the project will be made in the Institute in anticipation of the sanction of the scheme.

e. If the project involves the utilization of genetically engineered organism, it is agreed that we will ensure that an application will be submitted through our institutional bio-safety committee and we will declare that while conducting experiments, the bio-safety committee we will declare that while conducting experiments, the bio-safety guidelines of the Department of Biotechnology would be followed in toto.

f. If the project involves field trials / experiments / exchange of specimens etc. we will ensure that ethical clearances would be taken from the concerned ethical committees / competent authorities and the same would be conveyed to the Department of Biotechnology before implementing the project.

g. It is agreed by us that any research outcome or intellectual property right(s) on the invention (s) arising out of the Project shall be taken in accordance with the instructions issued with the approval of the Ministry of Finance. Department of Expenditure, as contained in Annexure - V.

h. We agree to accept the terms and conditions as enclosed in Annexure - IV. The same is signed and enclosed.

i. The Institute agrees that the equipment, the basic facilities and such other administrative facilities as per terms and conditions of the grant will be extended to investigators through out the duration of the project.

j. The Institute assumes to undertake the financial and other management responsibilities of the project.

1. Signature of Executive Authority of Institute with Seal and Date

2. Signature of Principal Investigator

3. Signature of Co-Investigator

4. Signature of Co-Investigator