RUBBER RESEARCH INSTITUTE OF INDIA

ANNUAL REPORT
2020-21

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Rubber Research Institute of India

The Rubber Research Institute of India (RRII), under the Rubber Board (Ministry of Commerce and Industry, Government of India), had its inception in 1955. With a very modest beginning, the RRII is now capable of handling most of the problems associated with natural rubber (NR) production technology, primary processing and product development. The steady growth of RRII in its scientific worth and research contributions has won it the recognition as an International Centre of Excellence in NR research.

Location

The RRII is located on a hillock 8 km east of Kottayam town in Kerala State and is easily accessible by road. Kottayam is connected to all major cities in the country by rail. There are two International Airports, one at Thiruvananthapuram, 160 km south and the other at Nedumbassery, 95 km north of RRII.

Organization

For the efficient discharge of its functions, the RRII has established major research divisions and research supporting sections at its headquarters and regional research establishments at appropriate locations where *Hevea brasiliensis* is commercially grown or is likely to be grown.

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ANNUAL REPORT
2020-2021

RUBBER RESEARCH INSTITUTE OF INDIA
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THE RUBBER BOARD

The Indian Rubber Board was constituted under the Rubber (Production and Marketing) Act, 1947, which came into force on 18 April 1947. This Act was amended in 1954, 1960, 1982 and in 1994. The Act was further amended by the Rubber (Amendment) Act, 2009 which came into force on 22nd January 2010.

Organization

The Chairman is the head of the organization. The Executive Director is the principal executive officer and exercises control over all Departments of the Rubber Board. The Research Department, Rubber Research Institute of India (RRII) works under the administrative control of the Board, the Director being the head of the Institute.

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Dr. Sawar Dhanania (upto 29.05.2020)

Dr. K. N. Raghavan IRS (From 30.05.2020)

Executive Director
Dr. K.N. Raghavan IRS

Secretary (In charge)
Smt. P. Sudha

Rubber Research Institute of India
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Director of Research

Crop Improvement
Joint Director (vacant)

Botany
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Principal Scientist (Officer-in-charge)

Biotechnology
Joint Director (vacant)

Dr. R.G. Kala
Principal Scientist (Officer-in-charge)

Genome Analysis
Dr. R.G. Kala
Principal Scientist (Officer-in-charge)

Germplasm
Joint Director (vacant)

Dr. Jayashree Madhavan
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Joint Director (vacant)

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Fertiliser Advisory Group
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Principal Scientist

Plant Physiology
Dr. K. Annamalainathan
Joint Director

Climate Change and Ecosystem Studies
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Crop Protection
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Joint Director (vacant)

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Scientist (Officer-in-charge)

Hevea Breeding Sub-station, Marthandam
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Instrumentation Officer (Officer-in-charge)

Library and Documentation Centre
Smt. N. Latha
Documentation Officer

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Sri. P. Aneesh
Assistant Statistition

Sri. K. A. Santhosh
Assistant Systems Officer

FOREWORD

RRII made some significant achievements during the year 2020-21 which included commissioning of the Rubber Products Incubation Centre, landslide vulnerability mapping of rubber plantations in Kerala, completion of the geo-referenced soil sample collection, analysis and fertility mapping of rubber plantations in north east India and developing a collaborative project with Spices Board and IIITMK for evolving an online fertilizer recommendation for cardamom in Idukki. Obtaining approval of Government of India to take up field trial of GM rubber in Assam was another important development in the reporting year. This assumes significance as more GM rubber plants are in the pipeline for which RRII would be approaching Government of India for permission to conduct their field trials.

RRII is also doing regular follow up studies on RubSIS in the traditional areas where this online fertilizer recommendation system has been in practice for the past couple of years.

A significant development has been the delinking of DRC testing Regional Laboratories from RRII and attaching them to the Rubber Board Companies. The original intention of the Regional Laboratories was to conduct soil testing but in recent years, these labs were acting mostly as DRC testing centres. The network of 7 DRC testing Regional Laboratories was insufficient to reach out to the nearly 1.2 million rubber growers in the country to test their DRC samples. Hence, Rubber Board Companies functioning in different parts of the country were assigned the duty of DRC testing to cater to a much larger section of the grower community.

In a situation where cultivation is expanding to non-traditional areas where cold,drought, high temperature, etc are major limiting factors, it is heartening to learnt the clone, RRII 430 is proving to be highly versatile, capable of adapting to these stressful environments. Poly cross breeding supported by Marker Assisted Selection and facilitated by the outcome from the whole genome sequencing project will go a long way to address the issue of potential yields reaching a stagnation and evolving high yielding clones more frequently. Tolerance to pests, diseases and climate resilience are increasingly relevant in the context of global climate change which is seriously impacting rubber cultivation in entire South East Asia.

Studies on cost of production of rubber in the major rubber growing states of the country, socio-economic impact of rubber cultivation in Tripura, adoption of good agricultural practices by rubber growers and studies on agrarian distress among small rubber growers are important projects that need to be addressed in a time-bound manner.

I congratulate the Director, Scientists and other officials of RRII for their service to the cause of natural rubber research. I recommend this report for perusal and study of all stakeholders in rubber value chain who will find the contents to be useful and interesting.

Dr. K.N. Raghavan
Chairman & Executive Director

DIRECTOR'S REVIEW

A few important projects with significant practical applications were completed during 2020-21. These included developing geo-referenced digital soil fertility maps of rubber plantations in Northeast India and landslide susceptibility maps of rubber holdings of Kerala, updating satellite-based geo-spatial mapping of rubber plantations in the entire country, assessing the agro-climatic suitability for natural rubber cultivation in different states, nutrient management of root trainer plants in nursery and monitoring changes in the quality of cup-lumps during storage. Commissioning a Rubber Products Incubation Centre at RRII was a major milestone of achievement during this year. With the soil fertility maps of Northeast India now completed, online fertilizer recommendations for the rubber holdings in the region can be launched. Assembly and annotation of *de novo* end-to-end whole genome sequencing of rubber is at the final stage.

It was evident from the geo-spatial studies that major changes were happening in the natural rubber landscape of the country. While the area under rubber cultivation in traditional regions has remained almost steady, cultivation has expanded in non-traditional areas, particularly in Northeast India. The geo-spatial technique employed to map rubber plantations will help monitor future changes in rubber cultivated area. Similarly, geo-referenced soil fertility mapping will help monitor long term impact of rubber cultivation on soil nutrient status.

Earlier studies have shown that India will need to increase domestic rubber production to achieve self-sufficiency and that this cannot be achieved unless there is a mission mode approach to replant the large share of old/senile rubber in the traditional region and expand cultivation to new areas in the non-traditional regions without causing significant adverse social or ecological effects. In this regard, the results of agro-climatic suitability analysis done for all districts in the country where rubber can be possibly grown assumes much importance. Next to traditional areas, productivity of rubber is likely to be the highest in Northeast India and current trends in global warming may bring more areas better suited for rubber cultivation in the region.

There have been efforts by block rubber processors to import cup-lumps to India which was welcomed by the tyre industry

and objected to by the growers. Our studies clearly show that upon storage the physical properties of cup-lumps deteriorate seriously, affecting their properties, making them a poor raw material for making good quality block rubber. Also, these cup-lumps harbor numerous microorganisms, maggots, etc. The COVID-19 pandemic and emergence of some major diseases in other rubber growing countries are strong pointers to the risks associated with importing unprocessed rubber. Processed rubbers such as block or RSS do not pose significant biological/ecological risks.

A few new projects were also initiated during this year. A new field trial was started at CES, Chethakkal incorporating five fruit crops by radically altering the planting geometry of rubber. There are discussions going on at the level of the state government in Kerala to allow cultivation of fruit crops in rubber estates. Outcome of this project will be of great practical significance for both small and large rubber growers. Studies on state-wise cost of production of rubber, socio economic impact of rubber cultivation in Tripura, adoption of recommended good agricultural practices by rubber growers and agrarian distress and livelihood issues of small rubber growers are other important new research projects initiated during this year.

RRII entered two new collaborations with two external agencies, namely (i) with Spices Board and IIITM-Kerala for developing an online fertilizer recommendation for cardamom in Idukki district of Kerala and (ii) with CSIR, NEERI to profile microbial population of rubber and forest soils through metagenomics analysis In addition, discussions are progressing with M/s. Michelin Tyre Company's rubber plantation division to extend R&D support which will include geo-referenced soil fertility mapping, satellite-based mapping of rubber holdings, inter cropping, etc. among others.

This Annual Report will be the final one that I am making as Director of RRII where I served 26 years, including more than 14 years as its Director. I was guided by the firm conviction that applied research should be strongly supported by the latest developments in basic science and technology to achieve the ultimate objective of science-society nexus. Keeping this in mind, new research avenues were opened in diverse fields of modern science and the results have been highly satisfying. Satellite-based geo-spatial techniques for mapping of rubber plantations in the country and geo-referenced soil fertility mapping, bioinformatics including end-to-end sequencing, assembly and annotation of the whole genome of rubber and marker assisted breeding, widening the genetic base of rubber by importing more than 40 best performing clones from nearly a dozen major rubber growing countries in the world, studies on climate change in the rubber growing regions of India and its potential impact on growth and productivity of rubber, ecological niche modelling, developing ICT tools for agricultural extension, etc. are a few such initiatives that have yielded tangible results and put RRII at a pre-eminent position among all rubber growing countries. Breakthrough innovations were made that made rubber cultivation less resource intensive and more efficient, sustainable and environment friendly. A record number

of high yielding clones were released, and their regional suitability was also recommended. Minimal cultivation practices were evolved that helped reduce cost of production, empowering rubber growers to withstand market volatility and instilling in them a renewed confidence in the future of rubber cultivation in the country.

Value addition during primary processing of latex and transfer of technology to the rubber products manufacturing industry were given much importance. Innovations led to improved product performance and reduced costs, energy consumption and CO_2 emission. Economic, environmental and social sustainability became the hallmark of Indian rubber industry.

Leading RRII has been a challenging but rewarding responsibility and a great privilege. I received the whole-hearted support and cooperation from the entire RRII fraternity which I gratefully acknowledge.

Dr. James Jacob

AGRONOMY AND SOILS DIVISION

Monitoring soil fertility status in rubber plantations and taking up research programmes to sustain soil fertility are important thrust areas of the Division. Generating additional income from the plantation through integrating diverse crops/ timber trees in rubber plantations is also a major area of research. With these objectives, various experiments on nutrient management and intercropping were continued. The homestead farming project with multiple enterprises initiated during 2019 was in progress. Follow up studies of RubSIS on liming and fertilizer skipping were in progress at different locations. Analysis of soil samples for fertility mapping of rubber growing regions of NE India was completed. Soil fertility maps were generated and development of online fertilizer recommendation for rubber growing regions of NE India was in progress. District-wise landslide susceptibility zones (shape files) available in the platform of Kerala State Disaster Management Authority (KSDMA) was used for the characterization of rubber plantations in Kerala according to landslide proneness. Rubber growing regions in Kerala were categorized in to low, medium and high landslide susceptible zones. A collaborative project was initiated with Spices Board and IIITMK for developing online fertilizer recommendation for cardamom plantations in Idukki district.

1. Nutrient management

The experiment on nutrient management initiated to revise the current fertilizer recommendation for root trainer nursery rubber plants was concluded. The third experiment, conducted including modified treatments based on the second trial also revealed the superiority of integrated management (25% of current fertilizer recommendation + *Pseudomonas* sp. + slurry, fortnightly application) followed by the treatment, foliar application of soluble fertilizers along with basal application of Mg and Ca.

The field experiment at CES, Chethackal to study the effect of long-term use of inorganic and organic manures on the growth and yield of rubber and on the physico-chemical properties of soil was continued without applying fertilizer and FYM to understand the residual effect of treatment.

The field experiment initiated during 2019 at TR & T estate, Mundakayam to study the effect of application of magnesium fertilizer on growth and yield of mature rubber was continued. Imposed treatments, recorded observations and compiled data.

Continued the Rhizosphere study and bulk soil samples were collected for the clones *viz.* RRII 430, RRII 414 and RRII 105 in four locations to identify a suitable clone in low pH soil and data analysis are in progress.

2. Intercropping and cropping systems

The field experiment at CES, Chethackal to find out the feasibility of establishing crops

like coffee (Selection 13), pepper, turmeric, amorphophallus, colocasia and chilli, as intercrops in mature rubber plantation under tapping was in progress. Planted ginger (IISR Varada) and three varieties of turmeric *viz*. IISR varieties, Prathibha, Pragathi and Alleppey Supreme as intercrops in mature rubber during 2020. Both the crops got established under the shade of mature rubber under tapping. The performance of ginger was poor as the crop was severely infected with soft rot. However, the performance of turmeric was very good and the yield realized was around 60 per cent of that of monoculture. Growth and yield of rubber did not vary significantly among treatments.

The experiment on Development of a multi species rubber-based cropping system for Tamil Nadu region was in progress. Growth of rubber continued to be significantly higher in intercropped area compared to control plot.

The experiments on intercropping perennial crops with rubber initiated at CES, Chethackal in 2001 were continued and the growth and yield of rubber continued to be not influenced by cultivating coffee or nutmeg as intercrops.

The homestead farming project was initiated at RRII during 2019. Rubber (clone RRII 430) was planted in paired rows at a spacing of 14x4x2.4 m. A control plot was also established at a spacing of 6.7x3.0 m. Perennial intercrops, mango, cashew and jack grafts were established as intercrops. Diverse food crops were planted in the interspaces. Pisciculture was also integrated in the homestead system. Girth of the plants were recorded.

The experiment initiated in 2016 in Malankara estate, Thodupuzha to study the effect of retaining pineapple after the intercropping period was continued. Growth and yield of rubber was not affected by retaining pineapple.

Table Ag. 1. **Effect of retaining pineapple on girth of rubber**

Year	Average girth of rubber trees (cm)		t-test
	Pineapple retained	Pineapple removed	
2020	70.06	68.81	NS

Experiment on inter planting of rubber with timber trees *viz*. teak, wild jack and mahogany was continued. Growth of rubber was not significantly influenced by row spacing and type of timber intercrops. No significant difference in yield of rubber was observed between spacing, types of intercrop and interactions.

To find out the feasibility of establishing fruit trees with rubber a field experiment was initiated at CES, Chethackal during 2020 and established rubber and exotic fruit trees (Mangosteen, Achachairu, Vietnam super early jack fruit, Abiu and Longkong) in the experiment area.

3. Ground cover management

The field experiment on the effect of legume covers and natural flora on growth of rubber and soil physico-chemical and biological properties laid out at the Central Experiment Station of Rubber Research Institute of India, was continued. The highest growth was recorded in *Pueraria* established plots followed by *Mucuna* and were comparable.

The study on differential effect of weed flora on growth of rubber and soil properties was continued. There was no significant difference between grasses and soft weeds on growth of plants. Dry weight of roots in the top 30 cm soil layer was higher for soft weeds.

The observational trial to find out the feasibility of establishing *C. caeruleum* under partial shade (after the intercrop pineapple) initiated at Kaliyar estate was concluded. *C. caeruleum* established well in the field and retained in the field in summer also. Based on the performance of *C. caeruleum* in initial and later immature phases, this is recommended as an alternate cover crop in rubber plantations.

4. Planting techniques

The experiment to evaluate different planting systems in 2007 at Cheruvally estate was continued. Canopy growth continued to be asymmetrical. Growth of rubber was comparable between control (square planting) and twin system of planting. In paired row planting, a strip of width 0.75 to 1 m was available without shading for intercropping. Yield of trees in the paired row system of planting was comparable with that of control (square system of planting).

5. Development of agro-management techniques for reducing the gestation period

The field experiment initiated at CES, Chethackal to develop an agronomic package to reduce the immaturity period of rubber starting from planting material onwards with combinations of two types of planting material and two management options as treatments was in progress. Direct-seeded green budded plants recorded significantly higher yield compared to green-budded stumps raised in polybags under current recommended practice.

The field experiment investigating the comparative performance of one- whorl, two-whorl and three-whorl polybag and root trainer rubber plants initiated at CES, Chethackal was continued. The yield did not vary significantly among different types of planting materials.

6. Stress management

For the drought study, rhizosphere the soil and leaf samples were collected from the clones RRII 414, RRII 430 and RRII 105 from the experimental field of RRII and analysis is in progress.

7. Soil fertility mapping and soil health monitoring

Follow up studies of RubSIS were in progress. The block trial at Thevervelil Estate, Perunadu, to study the effect of liming on growth of immature rubber and soil properties was continued. It was observed that liming had a significant influence on the growth of immature rubber. In the treatment with lime application @ 250 g plant^{-1}, more than 62 per cent of trees attained tappable girth in 67 months while in the control (no lime treatment) it was only 30 per cent (Table Ag. 2).

Table Ag. 2. **Effect of liming on girth of rubber plants**

Treatments	Status of trees after 67 months	
	Mean girth (cm) after 67 months	Trees with girth >50 cm (%)
Control	47.3	30
Lime @ 250 g plant^{-1}	50.5	62
Lime @500 g plant	47.8	42

The block trial at Thevervelil Estate, Perunadu to study the effect of liming on the yield of mature rubber and soil properties was continued. The dry rubber yield in the two treatments *viz.*, in lime applied and not applied blocks were significantly different.

Field experiments initiated during 2018-19 to study the effect of skipping of

fertilizers on growth and yield of rubber and on soil properties at Thevervelil Estate, Perunadu was in progress. The dry rubber yield in the fertilizer skipped and applied treatments were comparable. However, in the fertilizer applied plot significantly higher contents of soil available K, Ca and Mg were observed.

The experiment initiated at Malankara estate Thodupuzha as a part of the follow up studies to monitor growth and yield as well as soil and leaf nutrient status in rubber plantations following the *Adhoc* recommendation on skipping of fertilizers was continued.

The field trial to study the effect of skipping of fertilizers on growth and yield of mature rubber at Thirumbadi estate, Kozhikode was continued. Collected data on block yield.

Field experiements initiated at five locations in Kerala during 2018-19 and at one location in Karnataka to study the effect of skipping of fertilizers on yield and growth of rubber and on soil properties were in progress.

8. Soil fertility mapping of rubber growing regions of North East India

Soil fertility mapping of rubber growing regions of NE India was in progress. Georeferenced soil samples were collected from rubber growing regions of Tripura, Assam, Meghalaya, Manipur, Arunachal Pradesh and West Bengal and were analysed in the laboratory at RRII for different soil fertility parameters. Soil fertility status was mapped using geostatistics. Developing online fertilizer recommendation was in progress in collaboration with IIITM-K, Thiruvananthapuram.

8.1. Meghalaya

Natural rubber is cultivated in about 16330 ha in Meghalaya and its importance is growing in the State. Altogether 404 soil samples (0-30 cm) were collected from the State and subjected to chemical analyses for 13 parameters. From the rubber growing regions in the districts of North Garo Hills, West Garo Hills, East Garo Hills, South Garo Hills, South West Garo Hills and RiBhoi, 55, 162, 57, 39, 59 and 32 soil samples (0 –30 cm) were collected respectively.

8.1.1. Summary results

In general, about 35 per cent of the soils under rubber cultivation in Meghalaya state is categorized as extremely acidic. Most of the samples (> 98 %) analyzed were medium, high or very high in soil OC status. The same trend was observed in Av. Mg status also *viz.*, more than 98 per cent samples were medium or high. More than 85 per cent of the samples were either medium or high in Av. K and Av. Ca status. Among the macro nutrients, Av. P status was found to be deficient in almost entire samples. Deficient status of available S, B and Cu were found in more than 50 per cent of the samples while Av. Zn was sufficient in more than 75 per cent. Available Fe and Mn were sufficient in all the samples analyzed. Spatial variability of Av. Zn status is given in Fig. Ag. 1

8.2. Arunachal Pradesh

Arunachal Pradesh is primarily a hilly tract nestled in the foothills of Himalayas. Natural rubber is cultivated in about 5820 ha, spread over in six districts, *viz*, East Siang, Lower Siang, Lower Divang Valley, Lohit, Namsai and Papum Para and covers about 2.5 per cent of the total croped area of the state. Altogether, 92 soil samples (0-30 cm depth) were collected from the rubber growing areas and analysed for 13

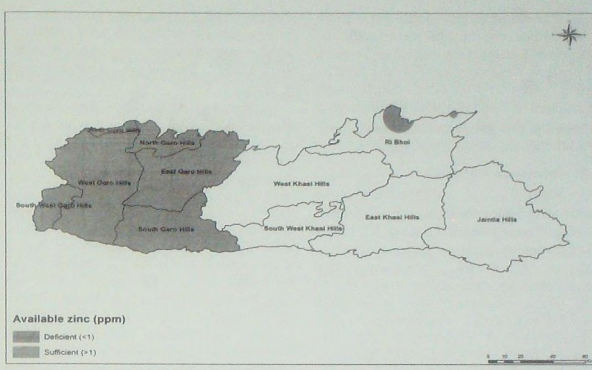


Fig. Ag. 1. Spatial variability of available Zinc in rubber growing soils of Meghalaya

parameters *viz.*, pH, organic carbon, exchangeable Al and available P, K, Ca, Mg, S, B, Zn, Cu, Fe and Mn.

8.2.1. Summary results

In general, the soils of rubber growing areas of Arunachal Pradesh are acidic in reaction, with eight per cent area in 'extremely acidic' category (3.5-4.5), 43 per cent in the 'very strongly acidic' category (4.5-5.0) and 39 per cent area in the 'strongly acidic' category (5.0-5.5) and 10 per cent area in "moderately acidic' category. Majority of the rubber area is medium in OC content with 63 per cent of the samples in 'medium' organic carbon category (0.75 – 1.5 %). OC was 'high' (1.5-2.5 %) in 28 per cent and 'very high' (>2.5 %) in 2 per cent area. Low organic carbon status (<0.75 %) was observed in seven per cent of the rubber area. About 51 per cent of the samples were low (<10 ppm) in available P content, while 38 per cent were in high (> 25 ppm) category and 11 per cent in medium (10-25 ppm) category. With respect to soil available K, 70 per cent of the samples were in medium category (50 – 125 ppm) while 25 per cent samples in low (<50 ppm) and 5 per cent in high (>125 ppm) category .

Majority of the area is high with respect to available Ca, with 75 per cent of the samples in 'high' category, and 12 per cent each in low and medium category. Available Mg is also high in general, with 82 per cent of the samples in 'high' (>25 ppm) category, and 16 per cent in medium (10-25 ppm) category. About 71 per cent of the samples were 'deficient'(<10 ppm) in av. S while 29 per cent samples were in "sufficient' (>10 ppm) category. Regarding micronutrient boron, about 66 per cent of the samples were 'deficient'(<0.5 ppm), while 34 per cent samples were "sufficient' (>0.5 ppm). With respect to micronutrient zinc, about 19 per

cent of the samples were 'deficient' (<1 ppm), while 81 per cent samples were "sufficient' (>1 ppm). As regards to available Cu, about 27 per cent of the samples were 'deficient' (<1 ppm), while 73 per cent samples were "sufficient' (>1 ppm). Entire rubber area in Arunachal Pradesh is in sufficient category with respect to available Fe and Mn.

8.3. Manipur

Natural rubber is cultivated in about 4200 ha in Manipur. Sixty seven soil samples (0-30 cm) were collected during 2019 from the rubber growing areas in the districts *viz.* Jiribam, Pherzawl, Temenlong and Tengnoupal based on satellite images during 2017. These soil samples were subjected to chemical analyses for 13 parameters *viz.*, pH, organic carbon, exchangeable Al and available forms of P, K, Ca, Mg, S, B, Zn, Cu, Fe and Mn.

8.3.1. Summary results

In general the soils under rubber in Manipur state were extremely acidic in reaction. About 82 per cent of the soil samples were extremely acidic (pH, 3.5-4.5) and rest of the samples were very strongly acidic (pH, 4.5-5.0). The soil pH varied from 3.75-4.96.

It was found that OC status of most of the samples were adequate whereas the entire samples were low in Av. P. About 51 per cent samples were medium (0.75-1.5%), 43 per cent of samples were high (1.5-2.5%) and 1.5 per cent of samples were very high (>1.5%) in OC status. The soil samples were low (<50 ppm, 54%) to medium (50-125 ppm, 46%) in Av. K status. About 63 per cent of samples were low in Av. Ca whereas 70 per cent of the samples were high in Av. Mg. High Av. Ca status (>150 ppm) was found in about 25 per cent of samples and 12 per cent of samples were medium. Most of the samples in the rubber growing area in this state were deficient in Av. S. Adequate level of Av. Zn, B and Cu were found in more than 50 per cent of samples. Av. Fe and Av. Mn were sufficient in these soils. Most of the soils were high to very high in Ex. Al.

FERTILIZER ADVISORY GROUP

The function of the group is to provide site-specific fertilizer recommendation on the basis of analysis of soil and leaf samples received from the large estates and small and medium growers. The laboratory at RRII headquarters is equipped with the facilities for testing the soil and leaf samples. The seven regional soil testing laboratories (RSTL) though initiated with the same purpose were mainly engaged in testing the dry rubber content of latex samples. Advices on fertilizer use were provided during the visit of the growers to the laboratory or as clarifications on telephonic enquiries or email queries. As a part of the restructuring of the Rubber Board, the seven regional laboratories were handed over to the Rubber Board managed Companies in the respective locality during the middle of August 2020. Details of sample analysis during the reporting year is provided in Table FAG 1.

Table FAG 1. **Details of soil, leaf and latex analyses**

Sample details	Number	Revenue (Rs.)
Soil	1823	4,25,526
Leaf	401	1,38,720
DRC of latex samples	12110	7,24,521
Total Revenue		12,88,767

CLIMATE CHANGE AND ECOSYSTEM STUDIES

This Division mainly studies climate change processes and its impact on rubber cultivation in traditional and non-traditional rubber growing regions of India. Another important area of study is developing information system on rubber cultivation using RS-GIS platform to account existing areas under rubber cultivation and identify suitable new areas where rubber cultivation can be extended. Meteorological data base management system in rubber growing regions is regularly updated and long-term data series are being used for climate change studies. Ecosystem level studies in rubber plantations like species diversity, microclimate parameters, effect of land-use change etc. are also being undertaken.

Geo-spatial mapping and updating of acreage of NR plantations in the traditional regions *viz.* Kerala and Tamil Nadu and in the non-traditional regions, Andhra Pradesh and Odhisha were completed during this reporting period. A total of 3.75 lakh ha of potential additional area for rubber cultivation was estimated in the non-traditional regions in India. Climate suitability for NR cultivation in the projected years 2050 and 2070 in 120 districts of NE India region was analyzed. Altogether, 105 districts could be identified in the Normal category followed by an additional 8 districts in the 10 per cent deviation category. For the 2070 scenario, a mean increase of 19 per cent higher favourability in climatic factors was predicted in NE states than for 2050 projections. Historical account of strong and weak episodes of el nino and la nina occurrences and their impact on rainfall pattern in NR growing regions of India was analyzed.

1. Developing rubber based information system using Remote Sensing and GIS

1.1. Geo-spatial mapping and updating of acreage using latest satellite data

1.1.1. Kerala and Kanyakumari district of Tamil Nadu

Updation of entire rubber plantations (aged 3 years and above) in the traditional rubber growing regions in Kerala and Kanyakumari district of Tamil Nadu was completed using satellite data as of March 2019. As per the satellite-based estimation, traditional rubber growing regions in Kerala and Kanyakumari district of Tamil Nadu had an area of around 606425 ha (Table CCES 1). Area under NR was the highest in Kottayam district (107708 ha) followed by Ernakulam (63073 ha), Kannur (55953 ha) and Pathanamthitta (54468 ha) districts. Around 47825 ha of area has expanded in this region between 2013 (satellite-based area) and 2019 and most of the area expansion occurred in the northern districts of Kerala. Out of the total expansion (47825 ha), around 65 per cent of area (31068 ha) increased between Trissur and Kasaragod districts. As of 2019, rubber plantations accounted for 15.3 per cent of the total geographical area and 22.6 per cent of the gross cropped area in the State. Critical interpretation of the satellite data revealed that replantation has taken place in many locations in Kanyakumari, Trivandrum, Kollam, Pathanamthitta, Kottayam, Idukki and Kozhikode districts. Spatio-temporal changes occurring in the rubber plantation landscape in the traditional regions is being regularly monitored.

Table CCES.1. **Satellite-derived acreage of NR (aged 3 years and above) in different districts of Kerala and Kanyakumari district of Tamil Nadu as of March/April 2019**

Sl. No.	Districts	Area under NR (ha)
1	Kanyakumari (TN)	21,933
2	Trivandrum	40,078
3	Kollam	46,935
4	Pathanamthitta	54,468
5	Alappuzha	8,490
6	Kottayam	1,07,708
7	Idukki	37,806
8	Ernakulam	63,073
9	Trissur	21,045
10	Palakkad	40,308
11	Malappuram	47,115
12	Kozhikkode	23,085
13	Wayanad	9,534
14	Kannur	55,953
15	Kasaragod	28,894
	TOTAL	6,06,425

1.1.2. Odisha

Mapping and updating of NR plantations in Odisha was completed. NR plantations were mapped using latest satellite data as of the year 2020 (Sentinel 2A/2B, March/April 2020). In Odisha, NR plantations are sparsely distributed in Mayurbanj, Baleswar, Cuttack, Gajapthi and Dhenkanal districts. Mayurbanj district is the dominant NR growing district in the State. Around 80 per cent of NR holdings are from this district. Satellite-derived acreage of NR in this State as of March 2020 was 681 ha (aged 3 years and above). Due to the scattered nature of holdings it would be important to update NR plantations in the State using high resolution satellite data.

1.1.3. East Godavari district in Andhra Pradesh

Updating and mapping of existing NR plantations in East Godavari district of Andhra Pradesh was completed. Satellite data as of April 2020 was used for the work (S2A MSI, April 2020). Around 339 ha of NR was estimated from the district (aged 3 yrs and above). Rubber plantations are sparsely distributed in Ramba Chodavaram and Addatigala taluks in East Godavari. Satellite-based estimation of NR area in this district as of 2012 was 239 ha showing an increase of 100 ha of area between the years 2012 and 2020.

2. Collaborative studies

2.1. Identification of potential areas for rubber cultivation in India

A Working Group (WG) for estimation of potential area available for rubber cultivation in India was constituted by Chairman and Executive Director of the Rubber Board in 2019. Accordingly, additional area available for rubber cultivation was estimated for different states in India for expansion of NR cultivation. The work has been completed and the technical report was submitted. As per the study the WG estimated an area of around 3.75 lakh ha in the country spread in the seven states of Northeast India (Sub-Himalayan states), three states in the Konkan region (Western Ghats) and three states in Eastern India (Eastern Ghats) for expansion of NR cultivation.

2.2. GIS based soil fertility mapping of NR in North-east India

As part of developing online fertilizer recommendation for rubber growing regions in North-east India (Rubber Soil Information System RubSIS), GIS based soil fertility mapping of Tripura, Assam, Meghalaya, West Bengal, Manipur and Arunachal Pradesh was completed. Soil fertility maps of Organic Carbon (OC), Soil pH, Available Phosphorous (P), Potassium (K) Calcium (Ca), Copper (Cu), Iron (Fe),

Magnesium (Mg), Manganese (Mn), Zinc (Zn), Sulphur (S) and Boron (B) were prepared in GIS platform. Soil fertility data sets of all these states (grid files of soil fertility maps, admnistrative boundary at district/taluk level) were structured according to the format of RubSIS (geotiffs and shapefiles) and transferred to Indian Institute of Information Technology and Management-Kerala (IIITM-K) for developing RubSIS platform. RubSIS platform was developed based on these maps for North-east India (https://rubsis.rubberboard.org.in). This was a collaborative work by RRII and IITM-K.

2.3. GIS database creation of panchayat level soil fertility status of rubber plantations

Developing GIS based panchayat level information on soil fertility status for NR growing regions in Kerala was completed. Geo-spatial data on soil fertility status (14 parameters) for 706 NR growing panchayats in different districts in the State and corresponding percent area were estimated. About 9884 grid files (of soil fertility) were generated for the work (706x14). Spatial information on this data would be useful to understand the soil fertility status of rubber plantations at Panchayat level. Besides, this outcome would act as a strong geo-spatial information on rubber soil information system at the local scale.

2.4. Delineation of landslide proneness of rubber plantations

In order to estimate the spatial extent of NR plantations according to proneness to landslide in Kerala, a study was initiated in September 2020. Satellite-based NR area (aged 3 years and above) was geo-spatially analysed with landslide susceptible zones of Kerala and the district-wise extent of NR susceptible to landslide (low, medium and high) susceptibility in the State were estimated. Results showed that out of the total rubber area in Kerala, 1.6 per cent (9485 ha) was in the high susceptibility zone, 6 per cent (32398 ha) in the medium and 2 per cent (13072 ha) in the low susceptibility zones (Table CCES. 2). More than 90 per cent of NR holdings are not situated in landslide prone regions. Area under NR in the highly susceptible landslide zone was the highest in Kottayam district followed by Idukki, Kannur, Palakkad and Pathanamthitta districts. This information is useful for planning appropriate conservation and management strategies for rubber plantations in the highly vulnerable areas. Rubber farmers can be better advised to mitigate the risks due to landslides.

2.5. Characterization of NR according to Agro-Ecological Zones (AEZs) and soil fertility status

Spatial extent of NR area according to different Agro-Ecological Zones (AEZs) in Kerala was analysed geo-spatially. Out of

Table CCES. 2. **The spatial extent of NR area susceptible to landslide in Kerala**

Sl. no.	Districts	NR area (ha) in the landslide susceptibility zones		
		Low	Medium	High
1	Trivandrum	162	12	3
2	Kollam	364	443	90
3	Pathanamthitta	3814	1668	512
4	Alappuzha	0	0	0
5	Kottayam	3266	7491	2371
6	Idukki	0	4961	2132
7	Ernakulam	299	900	65
8	Trissur	0	439	15
9	Palakkad	0	2479	909
10	Malappuram	0	2576	287
11	Kozhikode	0	2903	495
12	Wayanad	0	495	0
13	Kannur	0	7976	2121
14	Kasaragod	5167	55	485
	Total	13072 (2%)	32398 (6%)	9485 (1.6%)

24 AEZs in Kerala, the major share of area under NR was highest in the Southern and Central foothills (28%) followed by South Central Laterites (26%), Northern High Hills (12%), Northern Foot Hills (10%), Northern Laterites (8%), and Southern High Hills (7%). AEZs are delineated using altitude, rainfall pattern, soil type and topography. Thus the data can be useful for studying whether there are any differences in NR productivity according to different AEZs in Kerala.

Soil pH, Available Calcium (Ca), Available Potassium (K), Organic Carbon (OC) and Available Magnesium (Mg) were characterized geo-spatially according to different AEZs. Results showed that soil fertility status of the above parameters varied according to different AEZs, predominantly in the three zones *i.e.* South Central Foot Hills, Southern Central Laterites and Northern High Hills. The low status of Ca (<300 kg ha^{-1}) and K (<150 kg ha^{-1}) was exhibited in these three zones which accounted for around 50 per cent of the total NR area in Kerala. OC status was medium (22500-45000 kg ha^{-1}) and high (45000-75000 kg ha^{-1}) in these three zones which accounted for around 60 per cent of the total NR area in the State. Status of Mg was medium (30-75 kg ha^{-1}) and high (>75 kg ha^{-1}) and pH was extremely acidic (3.5-4.5) and very strongly acidic (4.5-5) in these three zones.

2.6. Developing GIS based soil fertility status of cardamom soils

In order to develop online soil fertilizer recommendation for cardamom plantations in Kerala, a consultancy project was initiated between Rubber Board, RRII and Indian Cardamom Research institute (ICRI), Idukki. As part of this programme soil sample distribution maps in Idukki district were prepared in GIS platform to check spatial distribution pattern of soil samples collected from different panchayats in Idukki. The map can be used as reference for soil sample collection in the cardamom growing regions of the district to collect grid-wise samples uniformly.

3. Identification of agro-climatically suitable areas for NR cultivation in India

In the context of fast changing climate scenarios, conditions affecting rubber growth and yield were analyzed for assessing the suitability of rubber cultivation for the immediate future and up to 2090 utilizing standard climate models for the Indian sub-continent. The methodology utilized for the current scenarios was adopted for the future model based global climate change scenarios through an appropriate General Circulation Model (GCM) for the rubber growing districts in the states of India. A GCM contains prognostic equations that are a function of time (typically winds, temperature, moisture, and surface pressure) together with diagnostic equations that are evaluated from them for a specific time period. The GCM which has been studied for the Indian subcontinent earlier, known as GFDL-ESM4 was adopted for the study. Data pertaining to rainfall and temperature was obtained for the projection years of 2050, 2070 and 2090. The data extraction was facilitated by utilizing the GIS software QGIS 3.6. These are considered as the mean year for periods 2040-2060, 2060-2080 and 2080-2100 respectively. The analysis was initially carried out for the NER with a total of 120 districts over 8 states of the NE Region.

The method of analysis for suitability was conducted by following two different analytical procedures. The first method employs the graphical area estimates of an

annual curve known as Area-Under-Curve (AUC) for each parameter. The AUC is defined as the total area under a curve in graph units. The agro-climatic conditions prevailing in Kottayam and Kanyakumari districts in the traditional NR growing region is generally considered as the most optimum and is adopted as the standard. Considering the total graphical area of these two districts separately as the benchmark for a particular climatic parameter, the extent of deviation in the climatic parameter of the non-traditional districts from Kottayam and Kanyakumari were determined. The percentage variations in AUC for each district from the respective chosen standard curves were calculated separately. Selection of a district was made if the AUC gives a 100 per cent or above match with that of either Kottayam or Kanyakumari district, which is considered here as the Normal category. Percentage deviation of AUC values from the standard, falling within the 10 and 20 per cent variability categories were also considered in the study. A district is not considered suitable if any one out of the six climate parameters fall beyond the 20 per cent deviation limit (tolerance limit defined for the purpose of comparison). The second method employs the "Crop Criteria" for growth and yield tolerance in terms of the Climate Tolerance Limits (CTL), which is defined as the extreme tolerance limit of important climatic parameters affecting growth and yield of rubber. Suitability of a district is determined only if it falls in either the AUC or CTL category.

A total of 120 districts were subjected to suitability analysis for the climate projection year 2050 based on the stipulated General Circulation Model (GCM) data of the NE region. Altogether, 105 districts could be identified in the Normal category (with that of Kanyakumari district) followed by an additional 8 districts in the 10 per cent category. Out of the 7 districts which were beyond the 20 per cent category, 6 districts belonged to the state of Arunachal Pradesh. For the 2070 scenario, a mean increase of 19 per cent is seen for the Aridity Indices which shows a higher favourability for 2070 compared to 2050 projections. The projected year of 2070 in the NE India revealed that 114 districts qualified for the Normal category where it is comparable to that of the Kanyakumari climate. The same number of districts holds good for the 10 per cent deviation and only a single district was added totaling 115 districts when the deviation was considered for 20 per cent category. Comparing the 2050 and 2070 projected years, climatic favourability of NE districts for rubber increased from 88 to 95 per cent in the Normal category which is comparable to that of Kottayam and Kanyakumari districts. The study will be extended to 2090 of the model data.

4. Characteristics of rainfall during 2020 for rubber growing regions in the traditional belt

Realized annual rainfall during 2020 was 101 per cent (3005.0 mm) of the normal in RRII, Kottayam. A near normal rainfall was experienced during all seasons with a total of 128 days. The SW monsoon rainfall amount showed an excess of 11 per cent from the normal, while that of the NE monsoon showed a deficit of 22 per cent. Peak rainfall activity was seen during the middle of first and second weeks of September, 2020. In CES, Chethakal, the annual rainfall received was 98 per cent of the normal (3332 mm) within 125 days. An excess of 121 and 111 per cent was observed during the Pre-monsoon and SW-monsoon periods respectively. One day highest rainfall was observed on 8th August 2020. The rainfall for RRS, Padiyoor amounted to

14 per cent (3718 mm) in excess of its long-term average. Excepting for the Pre-monsoon season which was deficient by 61 per cent, all other seasons received rainfall in excess of 83 per cent during the Northeast monsoon season. In Vaikundam Estate, Kanyakumari, the total rainfall was 2219 mm over 113 days. This was 115 per cent of the normal rainfall with a major contribution of 148 and 127 per cent from the Pre-monsoon and Monsoon rainfall respectively. The winter period did not receive any rainfall where an average of 3 days is usually received.

The Pre-monsoon rainfall received in the State was normal with a departure of 10.3 per cent from the normal. The actual rainfall received during the period was 404.7 mm. Pathanamthitta (856 mm) and Thiruvananthapuram (592.2 mm) districts received large excess rainfall during the period. The actual rainfall received in Kerala during the SW monsoon season was 2246.0 mm as against the normal rainfall of 2038.7 mm which was normal with a departure of 10.2 per cent from the normal. However, during the NE monsoon season 2020, the State received 314.9 mm of rainfall against normal rainfall of 445.8 mm which was deficient with a percentage departure of 29.4 per cent from the normal.

5. El Nino/La Nina episodes and regional climate in rubber growing regions of India

Monthly mean all India rainfall (1950-2015) was studied for periods of El Nino/La Nina. The monthly variations in the rainfall clearly pointed out that there was a deficit in rainfall during the El Nino year and a shift was observed in the subsequent year.

Mean monthly anomalies of rainfall were studied separately for all the monsoon months of El Nino and La Nina episodes for the year of occurrence and subsequent years. It was noted that rainfall was showing an inverse relation in the El Nino year. Excess rainfall could be observed when there was La Nina episode. The monthly values of Sea Surface Temperature (SST) anomalies and the rainfall anomalies revealed that maximum Nino 3.4 index occurred during 1982, 1997 and 2005. Maximum negative anomaly was observed during 1972, which was a strong El Nino episode.

Moderate El Nino episode in the year 2002 also showed negative anomaly. Maximum positive anomaly was found in the weak La Nina episode during the year 1983. Significant positive anomalies were observed in the La Nina episodes and negative anomalies during the El Nino episodes. During El Nino year rainfall was less in all the months during monsoon in the year of occurrence, whereas during subsequent year rainfall was less during June. Increased rainfall was observed during the rest of the monsoon months. Seasonal rainfall was low during El Nino and high during La Nina episodes during the year of occurrence. Reverse was the case for the subsequent years' rainfall. Above normal pre-monsoon rainfall was observed during La Nina episodes in the year of occurrence and subsequent years. During the subsequent year of an El Nino, rainfall increased from negative anomaly through June to September and further anomaly turning to positive till December. So it was clearly noted that there is a shift in rainfall pattern during the subsequent year of an El Nino episode.

During an El Nino period, rainfall is decreasing from strong to weak episodes during monsoon and *vice versa* in the case of La Nina episodes.

BOTANY DIVISION

Evolving high-yielding clones with resilience to biotic and abiotic stress is the prime mandate of the Botany Division. The main conventional breeding strategies being implemented include hybridization, ortet selection and polycross breeding. Evaluation of pipeline clones through a participatory plant breeding approach by involving estates and individual growers continued with an aim to identify region specific clones for large-scale commercial planting. Multi-locational clonal trials in zones limited by cold and drought stresses led to identification of pipeline clones with specific as well as wide adaptability. The studies also showed the versatility of the clone RRII 430 in terms of its superior performance in cold as well as drought prone regions. Breeding for disease resistance using disease-tolerant parents including other *Hevea* spp. resulted in identification of several outstanding progenies thus offering good scope for further long-term evaluation and subsequent adoption in disease-prone regions. Polycross breeding resulted in rapid recovery of several high-yielding selections. Studies were also initiated to investigate the utility of physiological, biochemical and molecular markers for early discrimination of high-yielders and stress-tolerant genotypes from seedling nursery evaluation stage onwards. Following international clone exchanges under the auspices of IRRDB, the *Hevea* gene pool in the country has already been augmented with high-yielding and disease-resistant clones from various countries. Steps were initiated to subject this new generation breeding population to physiological and pathological screening along with large-scale field evaluation.

1. Evolving high yielding clones for the traditional area

1.1. Hybridization and clonal selection

1.1.1. New generation hybrids

Hybridization programme involving different cross combinations were continued in the breeding orchard at RRII, Kottayam. Using 16 parental cross combinations, a total of 2591 hand pollinations were performed in 2021. A total of 784 seedlings which included 84 hybrids and 700 half-sibs were planted in seedling nursery 2020 for further evaluation. With an aim to develop high-yielding clones through transgressive segregation, hybridizations were carried out during 2011-14 using high-yielding Wickham clones (RRII 105, RRII 414, RRII 429 and RRII 430) as female parents and superior Wickham/Amazonian hybrids (95/10, 95/34 and 95/274) as male parents. Promising hybrids were selected for further evaluation under Clonal Nursery trial at CES Chethackal and the seedling nurseries were maintained at RRII. Promising half-sibs of RRII 430, RRII 414, RRII 429 and RRII 105 were selected for further evaluation under Clonal Nursery trial at CES Chethackal and the seedling nurseries were maintained at RRII.

1.1.2. Clonal nursery evaluation

Four clonal nurseries were planted in 2019 at CES, Chethackal using 268 selections which included 163 half-sibs, 75 hybrids, 24 polycross progenies and 6 ortets. Four check clones (RRII 105, RRII 414, RRII 417 and RRII 430) were included in the above trials. The above clonal nurseries were maintained and vacancies were also planted.

Out of 21 hybrid clones in clonal nursery (2008), six clones registered superior growth (02/688, 53 cm; 02/638, 48.8 cm; 02/690, 47.60 cm; 02/844, 53 cm; 02/514, 39 cm; 02/335, 52.3 cm) than check clone RRII 414 (34 cm). Based on yield over 4 seasons, top genotypes *viz.* 638 (26.7 g t^{-1} t^{-1}), 514 (24.3 g t^{-1} t^{-1}) and 335 (25.9 g t^{-1} t^{-1}) were selected. Bud wood nursery of the selected genotypes was established at CES, Chethackal and subsequently LST and PCE trials were concurrently laid out in four locations to evaluate their regional performance. Out of 26 clones in clonal nursery (2012), two clones *viz.* 05/139 and 05/432 continued to exhibit superior yield of 19.4 and 18.1 g t^{-1} t^{-1} respectively, than RRII 429 (13.0 g t^{-1} t^{-1}) and RRII 105 (10.1 g t^{-1} t^{-1}).

1.1.3. Small Scale Trials (SST)

Long term yield of 26 WxA hybrids in 1995B SST reaffirmed the superiority of five selections *viz.* 90/10, 90/271, 90/193, 90/170 and 90/17. Based on mean yield over 13 years, 90/10 was found the highest yielder with 72 g t^{-1} t^{-1} as compared to check clone RRII 105 (39 g t^{-1} t^{-1}) while 90/271 had more than 60 g t^{-1} t^{-1}, 90/193 and 90/170 had more than 50 g t^{-1} t^{-1}. Progenies of high-yielding family RRII 105 x RO 142 showed maximum resistance against ALF disease. Some of the selections are under participatory clone evaluation (Phase 4, 2014) in LST and OFTs.

Of the 24 hybrid clones tested in the SST (1999 C), five clones (94/87, 92, 296, 567 and 560) registered superior growth and yield (mean over nine years). These clones were pollarded and multiplied for further evaluation. In 2001 SST A, out of 25 clones, two were found superior to RRII 105 based on long term growth and yield. Hybrid clones *viz.* 95/413 (RRIM 600 x RRII 203) with 68.0 g t^{-1} t^{-1} and 95/425 (RRIM 600 x RRII 203) with 65.5 g t^{-1} t^{-1} continued to be superior when compared to RRII 105 and the better parent RRII 203. Among 29 clones in 2001 SST (B and C), two clones *viz.* 95/121 and 95/410 emerged superior to RRII 105. Of the 36 hybrid clones being evaluated in the SST 2003, two clones (96/417, 76 g t^{-1} t^{-1}; 96/459, 70 g t^{-1} t^{-1}) continued to register superior yield than RRII 105 (68 g t^{-1} t^{-1}), based on mean over seven years.

2. Evaluation of clones

2.1. Large scale evaluation (LST)

The LST (2019) of 14 clones (half-sib selections) was maintained at CES, Chethackal. In the LST of nine ortets from Cheruvally estate, five ortets showed significantly superior girth compared to check clone RRII 105 (Table Bot. 1). Mean yield of five ortet clones in BO-I panel was comparable with RRII 105. Although Cyo 72 was a superior yielder, it was found highly susceptible to ALF disease. Cyo 41, Cyo 35 and Cyo 48 are being evaluated in the PCE trials. Cyo 41 showed significantly superior girth, yield and very low incidence of ALF disease during the first two years in BO-2 panel.

2.2. On-farm evaluation

In the OFT of 14 pipeline clones at KFDC, Karnataka, three pipeline clones were superior to RRII 430, based on girth recorded in the sixth year of planting. In the trial of 12 monoclonal blocks, pipeline clones *viz.* P 70, P 21 and P 26 performed better in terms of girth (after three years of planting). Clones P 70 and RRII 429 recorded maximum girth (24.0 cm) and stem volume (0.01 m^3 each) followed by RRII 414 and RRII 417 (23.0 cm) and P 21 and P 26 (22.0 cm). Stem volume was 0.009 m^3 in clones RRII 414 and RRII 417 and 0.008 m^3 in P 21, P 26, RRII 422 and PB 330.

Table Bot. 1. **Girth and yield in ortets from HML Cheruvally**

Clone	Girth (cm)*	Mean yield (g t^{-1} t^{-1}) ALF incidence	BO-1 panel	BO-2 panel**
Cyo 41	79.1a	Very low	40.5ab	58.1
Cyo 18	66.5bc	Medium	36.9bcd	55.2
Cyo 43	64.4bcd	Medium	45.1ab	51.3
Cyo 30	69b	Low	46.0a	46.7
Cyo 35	67.7b	High	40.0abc	46.2
Cyo 48	82.1a	Low	31.2cd	43.8
RRII 105	59.4de	Medium	40.9ab	42.6
Cyo 72	62.1cd	Very high	47.6a	42.3
Cyo 31	64bcd	Medium	14.5e	25.3
Cyo 68	54.5e	Medium	28.2d	23.8

*Values followed by same letters are not significantly different based on DMRT; **2 years

3. Polycross progeny evaluation

In the observational trial (2005) at CES, Chethackal, polycross progenies had very high variability in terms of growth and yield. Girth during 15th year ranged from 34 to 130 cm and mean yield over seven years ranged from 10.8 to 99.4 g t^{-1} t^{-1}. Polycross progenies had a maximum mean girth of 86 cm as against 65.5 cm in clone RRII 105. Two polycross progenies had more than 90 g t^{-1} t^{-1} while one progeny had over 80 g t^{-1} t^{-1}. Five progenies had over 70 g t^{-1} t^{-1} and 12 had over 60 g t^{-1} t^{-1}. Sixty seven progenies had yield higher than the population mean (40 g t^{-1} t^{-1}) while 8 progenies yielded more than 70 g t^{-1} t^{-1} which was higher than the best yielding tree of RRII 105. These half-sib progenies with superior yield offer better scope for selection (Figs. Bot. 1a and 1b).

A polycross seed garden with 27 clones comprising of W x W and W x A hybrids, popular clones and germplasm accessions, *etc.* with a total of about 10,000 plants was planted in three experimental plots in an area of about 20 Ha at Arasu

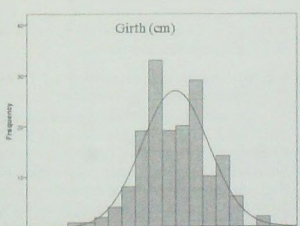


Fig. Bot. 1a. Frequency distribution of growth in polycross progenies

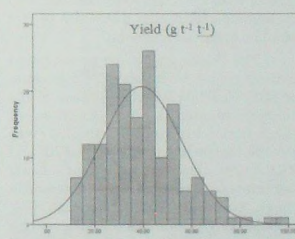


Fig. Bot. 1b. Long-term yield in polycross progenies

Rubber Corporation, Kanyakumari District of Tamil Nadu. During the year, a population consisting of 4400 polycross progenies was planted at Dapchari with 83 per cent survival. After completing one year of growth, seedlings attained an average height of 40 cm. In order to utilize the prepotency of PB clones and to develop high yielding clones through half-sib selection, 337 half-sib seedlings from high yielding PB clones were evaluated for yield by test tapping from which 70 half-sibs were selected and multiplied for evaluation under Clonal Nursery.

4. Genetic studies and investigations on genotype x environment interactions

4.1. Physiological evaluation of pipeline clones for drought and cold stress tolerance

With regard to the clonal nursery trial laid out during 2019 comprising 57 *Hevea* genotypes at RRS, Agartala and CES, Chethackal, influence of cold stress was assessed during February at RRS, Agartala. Effect of drought at CES, Chethackal was measured during March and the non-stress control measurements were also made on the same plants during the month of July after receiving sufficient rain.

The chlorophyll content was found reduced under the influence of cold stress at RRS, Agartala when compared to the control plants at CES, Chethackal. Maximum chlorophyll content was recorded in genotypes P 184, RRII 208 and RRII 429 under cold stress. Least degradation of chlorophyll was observed in check clone RRII 208. Though CO_2 assimilation (A), stomatal conductence (g_s), internal CO_2 (ci) and transpiration rate (E) got inhibited under cold stress across the genotypes, the instantaneous water use efficiency (inst WUE) was higher in cold stressed plants compared to control (Table Bot. 2). All the parameters showed a declining trend during March 2020 at CES, Chethackal when compared to stress free season (July 2020). Clones P 116 and P 93 showed maximum A and least photosynthetic reduction under low temperature indicating its cold tolerance potential. The check clones RRIM 703, RRIM 600 and RRII 429 also performed better in terms of A (6.5, 6.45 and 5.77 μmol m^{-2} s^{-1}) at Agartala whereas RRII 430 had lesser A (3.51 μmol m^{-2} s^{-1}). The genotypes with better A showed a general trend of relatively higher Gs, E, Inst WUE and lower Ci under cold conditions.

Under drought stress conditions of CES Chetheckal P 192 and P 197 maintained higher photosynthesis with lesser drought influenced reduction in photosynthesis. RRIM 703, RRII 208, RRII 414 and RRIM 600 maintained better A despite the stress condition. Most of the genotypes with a higher A tend to maintain higher E and low inst WUE which might be disadvantageous in terms of water utilisation. Only RRIM 600, RRII 414 and RRIM 703 were found to have comparable A with lesser trade-off on transpiration and high WUE thus confirming their inherent drought tolerance potential while RRII 430 displayed lesser A and higher transpirational loss.

4.2. Biochemical evaluation of latex parameters in clonal nursery

In the clonal nursery planted during 2012 both at RRS, Agartala and CES, Chethackal, biochemical parameters were analyzed in latex of 17 clones (selection of high and low yielders) during February 2020 at Agartala and May 2020 at CES, Chethackal. In terms of dry rubber yield, clones P 20, P 21, P 107 and P 57 performed better than other clones with 41.2, 37.6, 37.4

Table Bot. 2. **Net CO_2 assimilation rate (A), stomatal conductance (g_s), transpiration (Trmmol) and instantaneous Water Use Efficiency (inst WUE) of some selected *Hevea* genotypes at CES, Chethackal (March and July) and Agartala (February).**

GENOTYPES	A (μmol $m^{-2}s^{-1}$)			Gs (mol $m^{-2}s^{-1}$)			E			inst WUE		
	Agartala, Feb	CES, March	CES, July	Agartala, Feb	CES, March	CES, July	Agartala, Feb	CES, March	CES, July	Agartala, Feb	CES, March	CES, July
P 96	3.44	9.88	9.21	0.027	0.122	0.166	0.84	3.25	2.96	4.09	3.06	3.31
P 93	8.18	6.63	8.68	0.105	0.052	0.186	1.44	1.19	3.41	5.96	5.24	2.60
P 89	7.54	4.43	13.79	0.079	0.057	0.188	1.24	2.37	1.73	6.53	1.98	8.18
P 68	7.96	6.57	10.53	0.062	0.080	0.208	1.34	1.56	5.12	6.10	4.39	2.21
P 48	2.45	8.38	8.24	0.029	0.081	0.167	0.68	2.32	2.86	3.52	3.68	3.18
P 197	5.69	12.23	13.40	0.071	0.162	0.183	0.89	6.35	1.92	6.86	1.94	7.05
P 192	7.29	11.65	9.99	0.061	0.159	0.215	1.13	7.34	3.01	6.44	1.63	3.41
P 181	6.11	10.04	9.96	0.069	0.186	0.224	1.43	4.70	4.36	5.06	2.15	2.35
P 178	7.75	7.16	9.05	0.061	0.088	0.225	1.16	1.63	5.70	7.15	4.54	1.79
P 116	8.67	8.04	7.94	0.067	0.173	0.235	1.10	4.58	3.55	8.29	1.76	2.36
RRII 105	5.05	3.91	11.24	0.045	0.074	0.200	1.02	3.19	2.64	5.04	1.54	4.28
RRII 118	2.21	4.02	9.98	0.031	0.070	0.245	0.69	3.15	4.62	3.36	1.30	2.16
RRII 208	-	8.03	9.66	-	0.141	0.185	-	3.60	3.35	-	2.23	3.05
RRII 414	-	7.53	10.94	-	0.082	0.191	-	1.39	3.91	-	5.44	2.87
RRII 417	5.31	2.12	10.45	0.064	0.029	0.182	1.02	1.16	3.51	6.20	1.88	3.13
RRII 422	5.39	2.89	8.66	0.051	0.034	0.180	1.21	0.59	3.76	4.59	5.10	2.60
RRII 429	5.77	4.78	8.90	0.045	0.076	0.197	1.09	1.29	3.70	5.55	3.85	2.48
RRII 430	3.51	2.46	9.18	0.029	0.040	0.174	0.90	1.93	2.60	3.93	1.32	3.77
RRIM 600	6.45	7.50	12.18	0.08	0.081	0.205	1.27	1.26	4.76	5.92	6.07	2.64
RRIM 703	6.51	8.34	10.90	0.057	0.067	0.220	0.51	1.58	5.32	12.86	5.39	2.08

and 27.1 g t^{-1} t^{-1}, respectively while RRII 430 and RRII 105 yielded 20.7 and 19.2 g t^{-1} t^{-1}, respectively at Agartala (Table Bot. 3). In terms of ATP, the clones at Agartala were much inferior to plants grown at CES during March. Clone RRII 430 had the highest level of ATP in latex followed by P 110 and P 102 at CES. Other parameters such as sucrose, thiol and inorganic phosphate were also estimated and the clones were ranked based on rank sum method of all the parameters studied.

4.3. Study on relationship between concentration of ATP in latex and yield potential using a heterogeneous seedling population

In the study on latex [ATP] of seedlings test-tap yield ranged from 0.3 to 31 g $t^{-1}t^{-1}$, latex [ATP] ranged from 46 to 375 µM indicating the existence of very high variability for these traits in the experimental population. In general, high-yielding progenies had more latex [ATP] than low-yielders and *vice versa*. Regression analysis revealed a direct relationship (R^2=0.66) between latex [ATP] and test-tap yield of the progenies (Figs. Bot. 2a & 2b) showing that high test-tap yield and high latex [ATP] could ensure more precision in recovery of high yielding selections in nursery level.

4.4. Identification of pipeline clones with cold and drought tolerance based on growth and yield in clonal nursery

Analysis using the three environments as blocks showed that the environmental component accounted for major variation than others (clone, 19%; site/block, 65%; residual, 16%). Phenotypic coefficients of variation (*PCV*) were greater than the genotypic coefficients of variation (*GCV*) for growth and yield traits at all locations. Heritability for girth was very high (h^2 = 0.72) at Agartala while it was moderate at Chethackal (h^2 = 0.53) and Dapchari (h^2 = 0.41) (Table Bot. 4). Similarly, heritability for yield was very high at Chethackal (h^2 = 0.62), moderate at Agartala (h^2 = 0.42) and low at Dapchari (h^2 = 0.15). At Agartala, four pipeline clones *viz.* P 021, P 101, P 102 and P 107 possessed higher Performance Index (PI) than the cold-tolerant clone RRIM 600. At Dapchari, two pipeline clones *viz.* P 026 and P 102 had highest PI. Based on high PI under cold and drought stress conditions at Agartala and Dapchari respectively, P 102 emerged as a versatile pipeline clone with stable growth and yield under cold as well as drought environment. Three pipeline clones *viz.* P 021, P 101 and P 107 also possessed high PI displaying adaptability to cold and drought by sustaining superior growth and yield than RRIM 600 at both environments.

4.4.1. Identification of pipeline clones with ability to combine high yield and girth under cold and drought conditions

Results of correlation analysis indicated significant correlation between girth and yield. Among the three environments, correlation was maximum for Chethackal ($r = 0.77$, $p<0.05$) followed by Agartala ($r = 0.62$, $p<0.05$) and Dapchari ($r = 0.39$, $p<0.05$). In the cold-prone environment at Agartala, 17 pipeline clones *viz.* P 1, P 17, P 20, P 21, P 26, P 44, P 57, P 64, P 66, P 87, P 88, P 98, P 99, P 101, P 102, P 107 and P 110 along with RRII 430, RRIM 600 and RRII 105, could be identified as potential candidates for latex-timber clones since they were located in the quadrant for high yield and girth (Fig. Bot. 3). Under the drought-prone environment of Dapchari, 12 pipeline clones *viz.* P 20, P 21, P 26, P 44, P 60, P 61, P 64, P 70, P 101, P 102, P 107 and P 110 along with RRII 208 and RRII 430 were demarcated as potential latex-timber candidates with better growth as well as yield.

Table Bot. 3 **Comparison of girth, yield and biochemical parameters of selected *Hevea* clones experiencing cold stress at RRS, Agartala (RRS A) with stress free weather conditions at CES, Chethackal (CES)**

Clone	Girth		Yield		DRC		Volume		ATP		Sucrose		Thiol	
	CES	RRS A	CES	RRS A	CES	RRS A	CES	RRS A	CES	RRS A	CES	RRS A	CES	RRS A
P 10	43.6	26.68	32.46	11.46	41.58	43.79	80	27	355.7	79.1	11.08	1.88	0.223	0.297
P 101	39.9	35.48	33.26	18.73	36.15	43.13	94	43	353.0	78.1	14.50	2.98	0.299	0.293
P 102	38.4	42.05	20.16	12.63	32.19	35.23	64	37	532.8	129.6	7.35	1.81	0.250	0.531
P 107	30.4	45.01	18.34	37.35	35.80	44.82	56	83	204.3	81.2	13.54	6.53	0.317	0.305
P 110	37.7	43	21.55	22.48	38.02	46.09	60	50	544.8	95.6	11.78	6.5	0.267	0.397
P 15	38.6	26.40	39.42	12.77	32.68	38.75	121	33	233.9	93.9	4.43	1.32	0.284	0.377
P 17	43.8	35.54	54.49	22.08	35.86	42.03	152	53	367.5	159.1	12.03	10.99	0.351	0.303
P 20	34.63	37.24	22.63	41.22	37.93	48.88	66	83	214.7	118.5	6.90	7.99	0.277	0.303
P 21	20.25	47.24	12.95	37.64	32.17	45.39	40	84	152.3	171.8	10.63	7.98	0.317	0.364
P 57	27	36.82	11.19	27.10	33.77	43.26	32.5	62	265.0	90.6	6.17	2.97	0.240	0.331
P 62	28.9	29.80	26.00	5.81	33.54	44.11	77.5	13	206.8	39.6	7.35	4.16	0.274	0.317
P 63	39.6	32.55	26.48	9.05	37.54	36.42	70	23	348.0	86.9	5.47	2.2	0.264	0.471
P 66	23.6	28.30	13.39	8.76	33.77	47.08	40	19	315.8	91.2	4.78	3.58	0.223	0.475
P 84	28.9	32.16	21.47	8.67	38.44	48.19	56	6	204.4	89.2	10.79	5.74	0.324	0.374
P 9	33.63	24.43	17.67	9.19	28.92	47.70	60	19	112.9	81.5	6.97	5.74	0.289	0.355
RRII 105	23.7	29.05	15.03	19.24	33.09	44.92	48	43	361.0	79.0	9.86	3.6	0.272	0.369
RRII 430	39.7	37.44	41.09	20.70	37.95	45.49	108	45	572.6	108.6	7.67	5.98	0.167	0.352

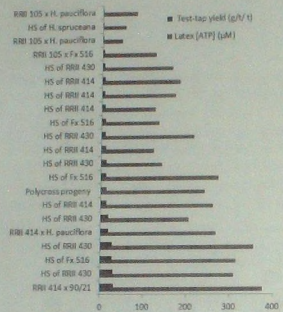


Fig. Bot. 2a. Relationship between latex ATP and test tap yield in seedlings

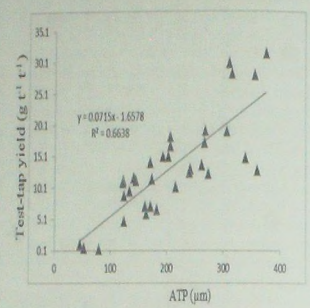


Fig. Bot. 2b. Regression analysis

5. Participatory evaluation of rubber clones in the pipeline

5.1. Source Bush Nurseries of pipeline clones

At the Central Experiment Station, Chethackal, 282 pipeline clones were maintained in 17 source bush nurseries.

5.1.1. Phase 1 (2008)

In LST 1, P 61 recorded a highest girth of 64 cm by 12 years. Clones P 10 and P 21 recorded girth on par with the vigorous check clone RRII 414 (60 cm). The girth increment rate at immaturity period of P 21 and P 61 was on par with RRII 414 (6.4 cm). Upon tapping, the clones P 10, P 60, P 61 and P 84 exhibited greater girth increment rate than the check clones. In the 4th year of tapping, clones P 60 and P 21 exhibited superior yield (67.3 and 58.2 g t^{-1} t^{-1}) than the check clone RRII 430 (42 g t^{-1} t^{-1}). In terms of leaf retention under ALF disease infestation, clones P 10, P 15 P 21, P 67 and P 84 were promising (Table Bot. 5).

In LST 2, P 63 recorded a highest girth of 63.7 cm by 10 years. Five clones registered vigorous growth comparable to RRII 414 (62.5 cm). The girth increment rate at immaturity period of P 70 and P 44 was comparable to that of RRII 414 (7.9 cm). Upon tapping, the clones P 62 and P 63

Table Bot. 4. Genetic parameters and heritability (h^2) of growth and yield of clones

	*GCV**	*PCV***	h^2		*GCV**	*PCV***	h^2
Girth				*Yield*			
Agartala	0.17	0.19	0.72	Agartala	0.21	0.32	0.42
Dapchari	0.13	0.21	0.41	Dapchari	0.18	0.46	0.15
Chethackal	0.13	0.18	0.53	Chethackal	0.41	0.52	0.62
Mean	0.14	0.19	0.55	Mean	0.26	0.43	0.40

*Genotypic coefficient of variation; **Phenotypic coefficient of variation;

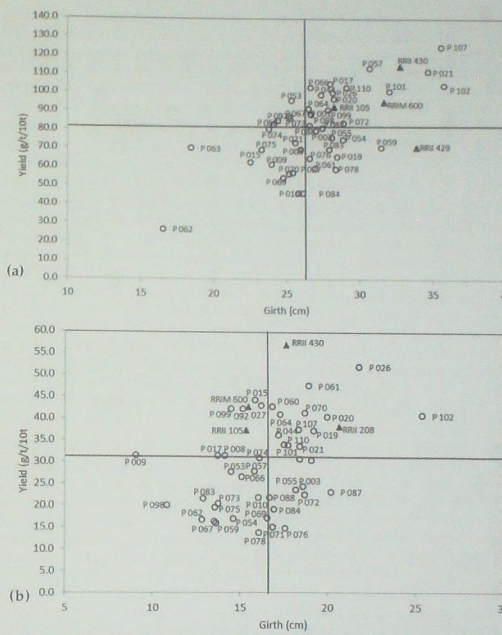


Fig. Bot. 3. Selection of pipeline clones with potential for timber and latex yield: (a) Agartala and (b) Dapchari

exhibited better girth increment rate than the check clone RRII 105 (2.4 cm). In terms of yield in the 4th year of tapping, clones P 26 and P 44 were superior (50 g t^{-1} t 1) and were comparable to the check clones RRII 414 and RRII 105. In terms of leaf retention under ALF disease infestation, clones P 62, P 63, P 64 and P 70 were superior.

In the OFT at Koottikkal estate (2008), growth was sub-optimal and ranged between 49.2 cm (P 62) and 62 cm (P 63 and RRII 414) by 12 years. Girth of P 26 was

Table Bot. 5. **Girth, girth increment (cm) and yield (g $t^{-1}t^{-1}$) of clones in PCE (2008) LST 1 and LST 2**

LST 1 Clones	Av girth (cm)	Rate of GI $year^{-1}$ (Immature)	Rate of GI $year^{-1}$ (Mature)	Mean yield (4^{th} year)	LST 2 Clones	Av girth (cm)	Rate of GI $year^{-1}$ (Immature)	Rate of GI $year^{-1}$ (Mature)	Mean yield (4^{th} year)
P 10	60.1	5.0	3.2	26	P-026	58.5	7.1	1.6	50.2
P 15	54.0	5.2	1.9	36	P-027	53.6	6	2.5	35.5
P 21	62.2	5.8	2.1	58.2	P-044	63.3	7.4	2.2	50.3
P 53	54.6	5.1	2.1	37	P-062	50.4	5.3	3.0	42.5
P 60	45.3	3.5	2.6	67.3	P-063	63.7	7.3	3.7	46.0
P 61	63.8	5.8	2.9	41	P-064	58.9	7.2	2.3	38.9
P 67	55.0	5.0	2.2	46.3	P-065	56.7	6.1	2.9	47.9
P 68	56.3	4.8	2.2	29.4	P-066	49.9	5.9	2.1	43.4
P 74	56.3	5.3	1.9	44	P-069	51.5	5.5	2.8	43.2
P 76	49.2	4.5	1.7	38.2	P-070	64.9	7.5	2.5	48.2
P 84	53.8	4.9	2.4	41	P-072	52.3	5.6	2	46.2
P 88	54.8	5.2	2.2	39.2	P-078	60.2	6	2.7	39.6
RRII 105	53.7	5.2	1.9	36	P-087	53.6	6.4	2.3	38.7
RRII 414	60.0	6.4	1.6	40	RRII 105	53.9	6.2	2.4	51.1
RRII 430	57.5	5.6	1.8	42	RRII 414	62.5	7.9	1.6	50.5
					RRII 430	58.9	7.1	1.9	48.7
CD 5%	3.60	0.73	0.58	11.6	CD 5%	3.4	0.4	0.7	9.0

comparable with that of RRII 430 (59 cm). RRII 430 had the maximum yield with 50 g t^{-1} t^{-1} over 3 years of tapping followed by P 66 with 43 g t^{-1} t^{-1}. Clones P 15 and P 26 were on par with RRII 105. Clone P 26 had more number of tapped trees and girth comparable to RRII 430 and DRC on par with RRII 105.

In OFT at Vithura, superior girth was observed in P 84 (70.7 cm) followed by P 61 (70.1 cm), P 68 (70.0 cm), and P 21 (70.2 cm) by 12 years. In the OFT at Devagiri Estate, Kanjirappally, P 26 continued to have comparable yield with the best performing check clone RRII 414. In the OFT at Athirapilly Estate, superior yield was observed in P 084 (67.2 g t^{-1} t^{-1}) followed by control clone RRII 414 (36 g t^{-1} t^{-1}). In the OFT at Be Be Estate Punalur, clone P 026, P 015 and P 066 registered superior yield when compared to RRII 430. In the OFT at Perinthalmanna, P 063 and P 087 exhibited superior growth and yield.

5.1.2. Phase 2 (2010)

The Central Large Scale Trial was planted in 2010 at Central Experimental Station, Chethackal with 14 pipeline clones and three check clones. Clone P 044 continued to exhibit superior girth and was comparable to RRII 414. Seven pipeline clones continued to maintain higher girth than RRII 105. Clones P 70, P 64, and P 19 continued to display lesser ALF disease incidence indicating their putative ALF tolerance. Preliminary assessment of yield over two years indicates RRII 414 (67 g t^{-1} t^{-1}) as the best performer followed by P 044 (65.3 g t^{-1} t^{-1}), P 093 (52.9 g t^{-1} t^{-1}) and RRII 430 (57.6 g t^{-1} t^{-1}). Three more pipeline clones performed better than RRII 105.

In the OFT at Vaniampara estate, four clones had a mean girth of above 60 cm as against 65 cm for RRII 430. Highest tappability of 89 per cent was observed in P 44 and P 54 among the pipeline clones. Yield over first eight months of tapping revealed clone P 64 as superior (52 g t^{-1} t^{-1}) followed by P 99 (50 g t^{-1} t^{-1}) while RRII 430 yielded 46.7 g t^{-1} t^{-1}. In the OFT at Kaliyar Estate, clone P 098 and P 099 registered highest yield. In the OFT at Pudukkad Estate, P 019 had maximum tolerance to ALF disease followed by RRII 430, P 047 and P 80.

5.1.3. Phase 3 (2012)

In the Central Large Scale Trial at Central Experimental Station, Chethackal, RRII 400 series check clones continued to occupy the top position in terms of girth thus substantiating their superiority. Six pipeline clones continued to retain better growth than RRII 105 among which P 110 and P 104 were toppers. ALF incidence was much lesser (< 10 %) in clones P 116 and P 156 and RRII 430 indicating their tolerance to ALF disease.

In the Chemoni OFT, clone P 110 recorded maximum girth (56 cm) followed by RRII 417 and RRII 430 (52 cm). Clones P 142, P 114 and P 104 had better girth than RRII 105 (47.8 cm). With regard to ALF disease tolerance, RRII 105 and RRII 417 had more than 95 per cent leaf retention while P 114 and RRII 430 had more than 80 per cent leaf retention indicating the putative tolerance in P 114.

In the OFT at Kumbazha estate, P 102, P 104 and P 110 were comparable to the check clone RRII 430 (57.6 cm) in terms of girth. RRII 430 followed by P 112 and P 104 registered superior yield in the first year. The DRC of P 104 was on par with RRII 430. At Calicut estate, superior girth was recorded in clone RRII 417 closely followed by P 110 and RRII 430. Most of the experimental clones were better than RRII 105 in terms of disease tolerance. Preliminary data on yield indicates RRII 417, RRII 430 and P 110 as superior performers.

5.1.4. Phase 4 (2014)

In the Central Large Scale Trial established during 2014 at RRII under Phase 4, three pipeline clones P 129, P 181 and P 172 continued to maintain better girth than RRII 105 and remaining clones. However, RRII 414, RRII 417 and RRII 430 continued to have superior girth in the trial. RRII 414 and RRII 430 had more than 90 per cent leaf retention displacing maximum tolerance to ALF. Among the remaining clones, P 129 followed by RRII 417, P 048, P 168, P 172 and P 180 had more than 80 per cent leaf retention. P 73 was the severely affected pipeline clone with less than 50 per cent leaf retention. In an on-farm trial at Chemoni Estate, four check clones attained maximum girth. Among the pipeline clones, P 168 and P 129 attained better girth. At the Kailiyadu OFT at Shoranur, P 171, P 129, P 168 and P 120 continued to display better growth after RRII 414.

At Chemoni Estate, P 121 exhibited better tolerance to ALF disease with more than 90 per cent leaf retention followed by RRII 417 and P 172. Severe infection was observed in P 73 with only 12 per cent leaf retention. In the OFT at Bethany estate, Kanyakumari, the trees were opened for tapping in the 6th year of planting. At Thiruampady estate, Kozhikode, four pipeline clones (P 168, P 133, P 126 and P 071) had better girth when compared to RRII 105 and RRII 414 though RRII 430 and RRII 417 were the top performers.

5.1.5. Phase 5 (2016)

In the 2016 OFT at Cherupittakavu Estate, Punalur, there are 14 pipeline clones and three check clones. Of these, RRII 430 continued to exhibit superior girth. Among the pipeline clones, P 200 and P 207 exhibited better girth than RRII 105. In the trial at SFCK, clone P 126 showed better initial growth (25 cm) than RRII 430 (23 cm). P 202 also showed early growth vigour. The plot of clone P 204 was fully damaged by fire outbreak. In the OFT at Paalali estate (Tamil Nadu), three pipeline clones exhibited girth superior to the top most check clone RRII 105.

5.1.6. Phase 6 (2019)

In the phase 6 of participatory clone evaluation trials (2019), casualty was assessed in the LST at CES and OFTs at ARC, Kanyakumari, SFCK, Punalur, RRS, Padiyoor and HBSS, Nettana. Initial establishment was better in Kanyakumari and Nettana. Replanting of casualties was undertaken in CES and Padiyoor.

6. Breeding for other specific objectives

6.1. Breeding for drought tolerance

In an attempt to develop drought tolerant clones for the non-traditional area, progenies developed through hybridization between high yielding clone (RRII 105; female parent) and a drought tolerant clone (PB 280; male parent) and the reciprocal crossing (PB 280 x RRII 105) were evaluated in a clonal nursery trial at RRS, Dapchari. In a trial with 40 experimental clones and nine control clones, six hybrid clones exhibited consistently superior juvenile yield than the check clone RRIM 600 in the peak yielding season (Table Bot. 6). The highest yielder was clone 114. Clone 69 was the second highest yielder which also exhibited highest girth. Three clones with superior yield performance in the peak yielding season exhibited superior yield during summer as well.

6.2. Breeding for disease tolerance

Introgressive hybridization was carried out between high yielding and

Table Bot. 6. **Girth and yield of top yielding clones**

Clone	Girth (cm)	Mean yield ($g\ t^{-1}\ t^{10}$)	Rank
114	17.9	71.7	1
69	21.6	69.5	2
66	18.4	67.7	3
59	15.4	62.2	4
117	17.2	61.1	5
98	18.8	60.9	6
RRIM 600	16.2	56.5	7
29	18.7	56.0	8
RRII 208	16.8	55.0	9
78	15.2	50.8	10
100	15.8	48.7	11
49	17.5	47.6	12
101	16.7	46.0	13
31	20.6	43.0	14
94	21.3	43.0	15
RRII 417	16.4	42.5	16
56	15.3	41.7	17
33	17.6	40.0	18
37	16.4	39.2	19
41	18.6	37.9	20

susceptible clones of *H. brasiliensis* as female parents and other disease-tolerant species (*H. spruceana* and *H. camargoana*) and wild accessions (RO 380 and RO 2871) as male parents. Half-sibs were also collected from a disease tolerant clone, Fx 516. Progenies were evaluated for juvenile growth and yield at the age of three years. Regarding growth in terms of girth, family RRII 430 x RO 380 (mean, 29 cm)and RRII 430 x *Hevea spruceana* (mean, 23 cm) performed well. Mean growth performance in families RRII 105 x *H. camargoana* and RRII 430 x *H. spruceana* was poor. With reference to juvenile yield, mean family yield of RRII 414 x RO 380 was 65 $g\ t^{-1}\ t^{10}$ followed by RRII 430 x RO 380 and RRII 430 x *H. camargoana* (15 $g\ t^{-1}\ t^{10}$), while the remaining cross-combinations produced progenies with low yield ranging from 7-9 $g\ t^{-1}\ t^{10}$. One exceptional progeny of the cross RRII 414 x RO 380 gave a very high yield of 191 $g\ t^{-1}\ t^{10}$. Half-sibs of Fx 516 had a mean yield of 17 $g\ t^{-1}\ t^{10}$ with maximum individual yield of 56 $g\ t^{-1}\ t^{10}$.

Preliminary assessment showed that the selections were putatively tolerant to major leaf diseases as there were no serious major disease symptoms. During the course of evaluation of above progenies, it was observed that many progenies were affected by a serious leaf disease associated with *Phyllosticta capitalensis* in the form of spots in mature leaves. Progenies of crosses *viz.* RRII 430 x RO 2871, RRII 414 x *H. spruceana*, RRII 414 x RO 380 and RRII 105 x *H. spruceana* had more than 80 per cent disease incidences in their progenies. Progenies of cross RRII 430 x RO 380 showed minimum disease incidence (I = 61%). Half-sib progenies of a disease tolerant clone Fx 516 showed lesser disease incidences (I = 25-27%) indicating better tolerance. In an on-farm trial, two clones *viz.* RRII 430 and RRII 417 recorded maximum (I = 60%) disease incidence. Clone RRII 422 (I = 10%) followed by RRII 414 (I = 20%) showed minimum disease incidence. Two other clones *viz.* RRII 105 and RRII 429 showed moderate incidence (I = 50%). Twenty five experimental clones of *Hevea* were investigated for their ALF disease incidence and corresponding leaf cuticular thickness which is primary frontline defense mechanism against plant pathogens. Using the above set of clones, a direct relationship was observed between incidence of ALF disease and leaf cuticle thickness ($R^2 = 0.53$), (Fig. Bot. 4). Above study indicated that thicker leaf cuticle might play an important role in imparting tolerance to ALF in *Hevea*.

The clones imported under international clone exchange from Vietnam, Thailand, China, Cambodia and Indonesia including SALB-resistant clones, were assessed for putative resistance to major fungal pathogens in collaboration with

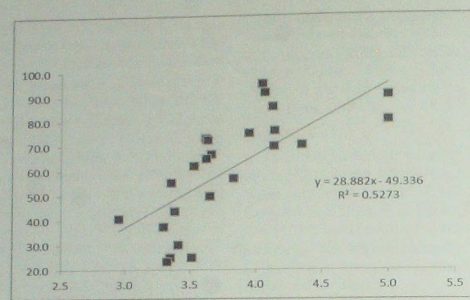


Fig. Bot. 4. Relationship between leaf cuticle thickness and ALF disease in 25 genotypes of *Hevea*

Pathology Division. The clones imported from Vietnam, Thailand, China and Cambodia were found to be variably tolerant to dreadful fungal leaf pathogens like *Corynespora*, *Colletotrichum* and *Phytophthora*.

7. Molecular breeding

7.1. Abiotic stress responsive SSR markers

This study is aimed to employ SSR markers identified from abiotic stress responsive transcripts of *Hevea* for construction of genetic linkage map in *Hevea*. Arrangements were made to get the genomic DNA isolated from the selected genotypes for PCR analysis. The work is in progress.

7.2. Validation of molecular markers for abiotic stress tolerance

This study was initiated to validate the molecular markers developed from previous studies on abiotic stress responsive and yield related transcripts of *Hevea*. Primers for these markers were designed and synthesized. Leaf and latex samples were collected from drought and cold stress exposed plants of about 50 genotypes grown at RRS, Dapchari and RRS, Agartala, respectively and from CES, Chethackal (control) and stored frozen for further downstream processes.

8. Anatomical investigations

8.1. Petiolule anatomy for characterisation of *Hevea* clones

Anatomical characterisation was carried out using indigenous and exotic clones which included 43 clones imported from 10 countries under bilateral clone exchange programme of the IRRDB. Characterisation of these clones is a mandate of this programme and is being carried out in the country. The study on distal pulvinus of *Hevea brasiliensis* leaves revealed the existence of maximum diversity for structural traits with reasonably good stability for characterisation of clones from

Cambodia and Thailand. Possibility of using structural traits *viz.*, inter-vascular continuity, shape of vascular bundles, xylem characteristics and proportion of tissues in the stele for characterisation of these clones was also investigated. A systematic key for the identification of the above mentioned clones was formulated based on structural and morphological traits.

Table Bot. 7. **Effect of age of bud wood stock on tappability and yield**

Treatment		Tappability (%)	Mean yield over six years ($g\ t^{-1}\ t^{-1}$)
T1	20 years nursery A	59.5	60.0
T2	4 years nursery A	59.3	58.3
T3	4 years nursery B	70.7	56.4
T4	4 years nursery C	51.3	60.0
T5	20 years nursery C	38.5	62.4
T6	10 years trees*	8.0	53.8
T7	1 year nursery B	59.3	60.3
	CV	—	8.5
	LSD ($p<0.05$)	—	6.0

* Buds collected directly from the terminal branches

9. Studies on propagation

9.1. Studies on effect of age of budwood stock and quality of planting materials

The study on effect of age of bud wood stock and production of quality planting material over six years revealed that the age (12-20 years) of bud wood stock does not influence the quality of the planting materials (Table Bot. 7). However, use of buds directly collected from field trees resulted in trees with very low tappability and yield.

In another study, type of bud did not influence the quality of planting materials and there was no significant difference between treatments with respect to trunk growth and initial yield (Table Bot. 8).

Table Bot. 8. **Effect of type of bud source on growth and yield**

Type of bud source		Girth (cm)	Mean yield over 6 years ($g\ t^{-1}\ t^{-1}$)
T1	Brown	58.0	51.0
T2	Conventional	59.4	49.8
T3	Semi-green	54.3	50.0
T4	Scale bud	54.0	46.2
T5	Whorl bud	58.6	48.2
T6	Light green	60.2	49.5
T7	Unhealthy buds	57.9	47.0
	CD	NS	NS

9.2. Role of sprouting pattern of seeds on seedling growth in *Hevea brasiliensis*

A study was carried out to assess the influence of sprouting and early shoot and root characteristics of *Hevea* seedlings on seedling growth and uniformity. Germinated seeds were grouped into four categories based on the shoot and root length characteristics and planted in polybags. After three months of growth, buddability and shoot-root length were recorded. Maxmimum buddability (76%) was observed in seedlings grown from germinated seeds with lengthy shoot and root. Germinated seeds with short shoot and root length had minimum buddability (30%) and lesser uniformity in plant growth. The above plants were transplanted to polybag for field planting.

10. International clone exchange

10.1. Large scale trials

SBNs of the imported clones from Sri Lanka were established at CES, Chethackal. Six imported clones from Cote d'Ivoire and

Myanmar were bud-grafted for the proposed field trial. Two large scale trials (LST) established at CES, Chethackal with clones imported from Thailand (RRIT 226, RRIT 251, RRIT 408, RRIT 3604 and RRIT 3904), Indonesia (IRR 5, IRR 104 and IRR 119), China, Vietnam and Cambodia are continued. SBNs of the imported clones from ten countries including Thailand, Indonesia, Philippines and Sri Lanka, etc., are also maintained at CES, Chethackal. The above clones are being screened for their abiotic and biotic stress tolerance using laboratory experiments in collaboration with Pathology and Physiology Divisions.

10.2. International *Hevea* Clone Museum

The International *Hevea* Clone Museum, consisting of high-yielding as well as SALB-resistant clones imported from different IRRDB member countries is being maintained at RRII main campus. In collaboration with Pathology Division, the imported clones were screened for major fungal disease pathogens through laboratory bioassay and clones with putative tolerance were identified.

11. Arboreta of *Hevea* clones and forest species

The arboretum of *Hevea* consisting of 55 clones which serves as demonstration-cum-research plot, was maintained at RRII main campus, Kottayam. Similarly, an arboretum of 63 assorted forest plant species along with *Hevea*, planted in the RRII main campus during the year 2014 is also being maintained.

12. Upgradation of Prang Besar (PB) clones to Category 1 and release for cultivation in traditional region

The upgraded clones *viz*. PB 280, PB 255 and PB 314 were multiplied and the root trainer plants are being maintained for establishing more source bush nurseries.

13. Supplementary research for clone evaluation trials

13.1. Quick coagulation for sheet rubber processing

In continuation of the method devised for DRC estimation of *Hevea* latex, a new methodology was developed for quick coagulation of latex for the preparation of sheet rubber. Raw rubber properties and technological properties of sheet rubber from quick coagulation were comparable with the standard protocol (Tables Bot. 9 and 10).

Table Bot. 9. **Raw rubber properties of sheet rubber from standard and rapid coagulation**

Parameter	Standard protocol	Rapid coagulation protocol
P_o	29	28
PRI	90	82
Mooney viscosity (ML(1+4) 100°C)	54	54
Colour	Honey colour	Honey colour

Table Bot. 10. **Cure characteristics of sheet rubber from standard and rapid coagulation**

Sample	Cure characteristics				
Particulars	$S'M_L$ dNm	$S'M_H$ dNm	Tan δM_L	Tan δM_H	T90 (min)
Standard	1.13	14.43	0.88	0.05	6.34
Rapid	1.29	15.15	0.83	0.05	6.27

GERMPLASM DIVISION

The *Hevea* germplasm maintained at RRII includes the domesticated gene pool with clones derived from the original Wickham collection of 1876, the wild germplasm belonging to the 1981 IRRDB collection, and the collection of other *Hevea* species. Maintenance of the domesticated genepool collection, introduction and conservation of remaining *Hevea* species, conservation of the wild germplasm, its agronomic evaluation, screening for diseases, drought and cold stress resistance, timber latex traits and molecular characterization are the major activities of the Division.

1. Introduction, conservation and documentation

1.1. Domesticated gene pool (Wickham collection) from secondary centers

The 183 Wickham clones belonging to this genepool are being conserved in a budwood nursery (the clone museum) at RRII, Kottayam, and three arboreta (germplasm gardens) at CES, Chethackal. The arboreta serve the primary purpose of conservation, scientific data collection as and when necessary, and are a source of clonal flowers for breeding when required.

51 Wickham clones are being conserved in an arboretum planted originally in 1977, and rejuvenated by ratooning in 2000. Wintering pattern was recorded in these clones to characterise early and late wintering clones.

Table Ger. 1. **Mean yield of IRCA clones over 19 years**

Clone	Mean yield (g t^{-2} t^{-1})
IRCA 130	62.24
IRCA 109	52.34
IRCA 111	61.30
IRCA 18	55.09
IRCA 230	53.95
RRII 105	53.84

Long term evaluation of the five IRCA clones introduced in 1991, showed that IRCA 130 was the best latex timber clone, followed by IRCA 109 (Table Ger. 1). These clones have been upgraded to Category 2.

In an arboretum comprising 20 Wickham clones in Germplasm Garden 94, RRIM 609, RRIC 100 and RRII 23 showed the highest average yield over 16 years of tapping (79.0 - 75.5 g t^{-2} t^{-1}), compared to 50.8 g t^{-1} t^{-1} for the popular clone RRII 105. RRII 23 also continued to be the most vigorous clone, and had the highest girth increment rate (86.2%) after tapping, compared to RRII 105 (53.4%).

1.2. 1981 IRRDB wild germplasm

This gene pool was introduced during 1984-1990 into the country, and is the focus of the division.

1.2.1. Conservation nurseries

The original nurseries established at the time of introduction, continue to serve as a source of flowers for hybridization programmes. These accessions were re-established in compact new conservation-cum-source bush nurseries (SBNs) from 2003 to 2008. 255 potentially useful accessions identified over the years during the characterization and preliminary evaluation in the juvenile stage for yield, yield contributing traits like latex vessels, disease and drought tolerance traits in these SBNs, are being put into Further Evaluation Trials in phases. Simultaneously, these selections are also being established in separate bud wood nurseries (the Germplasm Working Collection) in order to ensure better care and accessibility. So far,

119 potential wild accessions have been established in the GWC, to which potentially useful accessions will be added as and when identified.

1.2.2. Heveatum

A separate *Hevea* arboretum (or *Heveatum*), comprising all the available genetic resources, is being established in phases at Teksragre farm, Tura, Meghalaya, with the primary intention of ensuring an insurance collection, as well as facilitating free cross pollination and genetic mixing between the different gene pools. Multiplication for the next phase could not take place due to covid constraints. So far, a total of 577 wild and Wickham accessions have been established here in five phases. Another arboretum comprising 120 accessions established earlier, is being maintained at Central Experiment Station, Chethackal.

1.3. Other *Hevea* species

This collection comprises five species other than *H. brasiliensis* (*H. benthamiana, H. spruceana, H. nitida, H. camargoana* and two accessions of *H. pauciflora*) and are being conserved as an arboretum established in 2006 at CES. Five natural putative interspecific hybrids, two *H. brasiliensis* clones, and FX 516 (an interspecific cross between *H. brasiliensis* and *H. benthamiana*) have also been planted here. Among the other species, *H. pauciflora* from Sri Lanka, as well as Fx 516 continue to show relatively good yield.

2. Characterization and preliminary evaluation

The last three trials in the Preliminary Evaluation Trial format were planted at RRS, Padiyoor in 2000 (A&B) and 2002. Nine relatively high yielding selections and five vigorous accessions for timber traits are being conserved as male parents for future W x A hybridization programmes.

3. Further evaluation and selection

Selections from preliminary evaluations with 50-80 per cent of the test tap yield of the controls, are evaluated in detail in clonal nurseries (CNs), and those with more than 80 per cent test tap yield of RRII 105, in field trials (FETs) at normal spacing.

3.1. Clonal nursery evaluation

The clonal nursery planted with 15 accessions. had identified 3 potential accessions of which one has been taken to FET 2019 in Nettana for further evaluation.

3.2. Further evaluation trials

All accessions with more than 80 per cent of the control yield on preliminary evaluation are subjected to detailed evaluation in FETs in statistically laid out trials at normal spacing. There are currently eight FETs comprising 150 accessions, including two FETs planted at CES and Nettana, last year.

Twenty-two wild accessions and three control clones are being evaluated for growth and yield traits in the FET 2003. Accession RO 2629, AC 4149, and AC 716 continued to record the highest yield, while RO 2629 followed by MT 2233 and AC 626 recorded the highest girth. Among the 22 wild accessions in FET 2005, AC 2004 and MT 43 were the most vigourous (girth of 83 and 80.2 cm respectively), while the controls PB 260, RRIM 600 and RRII 105 had girth values of 77.7, 66.1 and 58.1 respectively. RRII 105 had the highest yield over 5 years of tapping, followed by MT 4788, PB 260, AC 2004, RRIM 600 and MT 43. Significant clonal differences were seen for the trait

Number of Latex Vessel Rows (NLVR). MT 4788 had the highest NLVR of 30.5, compared to 19.7 of RRII 105. MT 2217 which had been originally selected from a PET for its high NLV, was on par with RRII 105 with 18.3. The NLVR for controls PB 260 and RRIM 200 were 14.3 and 12.5 respectively. In FET 2008 comprising 26 wild accessions, RO 2846, AC 176 and MT 200 were the most vigourous. Among the 13 accessions evaluated in FET 2010 at CES, Chethackal, there were seven accessions with girth higher than clone RRII 105 and none of the accessions were superior to check clones RRII 430 and RRII 414. In FET 2013 comprising 22 selected wild accessions along with three control clones in the seventh year of growth at CES, growth was monitored. Wild accessions AC 167 , AC 5280 and RO 2784 had the highest girth. Two more FETs planted in 2019- FET 2019a and b comprising 23 and 10 wild selections, are under evaluation at CES and Nettana respectively in the second year of growth.

3.3. On-farm trials

Selections from FETs are subjected to multi location evaluation in On-Farm Trials for confirmation of yield potential. Of the first OFT established in 2010 at five locations *viz.* B.C. Cheruvally estate in Erumely, Malankara estate in Thodupuzha, Mooply estate in Thrissur, Calicut estate in Kozhikode and Bethany estate in Kanyakumari for evaluating the performance of the three selected IRCA clones (IRCA 130, IRCA 111, IRCA 109) and one wild accession (AC 166), one location was lost in 2017 due to cyclone Ockhi. Yield recording in these estates was disrupted due to Covid this year.

4. Screening for stress tolerance

4.1. Screening for biotic stress tolerance

Forty one short listed wild *Hevea* accessions along with two control clones are under evaluation for confirmation of field tolerance to *Corynespora* disease at Ullickal Nursery, Iritty.

4.2. Abiotic stress resistance

4.2.1. Drought tolerance

Twenty half-sib progenies selected from 40 half-sibs of nine clones in a Clonal Nursery evaluation at RRS, Dapchar, and six out of 31 half-sibs from three pre-potent clones in a CN at RRS, Padiyoor were maintained in a bud wood nursery for further multiplication for advancing to a large scale trial at RRS, Dapchari for developing location specific clones.

In the further field evaluation of selected *Hevea* clones at RRS, Dapchari in collaboration with Botany Division, yield in the 34 selected *Hevea* clones planted in 2007 comprising 23 wild accessions, five HP clones and six check clones *viz.* RRII 430, RRII 414, RRII 105, RRIM 600, RRII 208 and Tjir 1 was assessed. Among the wild accessions, highest mature yield was recorded by MT 4788 (22.6 g $t^{-1}t^{-1}$) followed by MT 4856 (14.4 g $t^{-1}t^{-1}$). Four other wild accessions also were found to be promising. Among the hybrid clones, 93/105 recorded highest mature yield of 25.1 g $t^{-1}t^{-1}$ .The check clone RRII 430 recorded highest yield among all (50.1 g $t^{-1}t^{-1}$) proving its drought tolerance. Highest girth was recorded by accession MT 1619 (58.3 cm.) followed by MT 4788 (551.7 cm.). Out of five hybrid clones evaluated, hybrid 93/105 recorded highest girth (51.0 cm.) and the highest girth among check clones was in RRII 430 (55.1 cm.)

4.2.2. Cold tolerance

Two trials comprising of 64 wild *Hevea* accessions along with check clones are under evaluation for cold tolerance at the

Regional Experiment Station, Nagrakata, West Bengal. Highest girth was recorded in the accession RO 2902, MT 923 and MT 5105 as compared to the check clones SCATC 93/114 and RRIM 600 in Trial 1, while accession RO 2727, MT 915, and RO 3197 recorded the highest girth compared to that of the controls Haiken 1 and RRIM 600 in Trial 2. Among the wild accessions studied, AC 4653, AC 3514 and MT 915 recorded the highest yield. Selected wild accessions were included in the abiotic stress tolerance studies of Plant Physiology Division.

5. Screening for timber characteristics

5.1. Field screening

Three wild accessions MT 941, MT 1032 and AC 650 had high timber potentiality out of 25 genotypes being evaluated for growth at RRS, Padiyoor.

6. Utilisation of *Hevea* germplasm

6.1. Hand pollination programmes

Hand pollination programmes involving potential wild accessions and selected elite Wickham clones commenced 2009 for broadening the genetic base as well as genetic improvement of the crop.

The HP 2009 hand pollination programme at CES, Chethackal, involving three wild accessions and six cultivated Wickham clones, generated 75 WxA progenies along with 70 OP seeds, of which six hybrids and one OP seedling were promising. The HP 2009 hand pollination programme at RRS Padiyoor, yielded 10 promising hybrid progenies from two parental combinations, and seven OP seedling progenies of RRII 105, which are now in the second year of clonal nursery evaluation at HBSS, Nettana. The WxA and interspecific (RRII 105 x *H benthamiana*) HP programmes at CES and RRII from 2009 to 2014 generated 457 progenies, of which 33 were very promising and have are being further evaluated in an FET 2019 at CES.

In the 2016 HP, of the 75 surviving W x A hybrids from three W x A cross combinations, progenies of the cross with the *Oidium* tolerant parent RO 2871 showed the highest vigour and survival percentage. The second round of test tapping confirmed the potential of the two high yielders identified last year (#44 and 90), along with 9 other medium yielders, and will be taken up for further evaluation next year.

A total of 45 potentially high yielding WxA progenies out of 222 generated have been obtained since 2009, of which 30 most promising ones have been taken up for further evaluation in 2019. Among the interspecific progenies, 71 out of 255 were promising, of which 13 of the most promising have been taken up for further evaluation in 2019.

In the 2019 HP, 48 hybrids obtained were planted in the seedling nursery for evaluation. In 2020, 1100 crosses between eight selected wild accessions and three Wickham clones were made, with an initial fruit set of 58 (5.3%): however only 9 seeds could be salvaged finally since timely prophylactic measures could not be taken due to COVID restrictions. This flowering season too, initial fruit set of 82 was obtained from 1036 crosses WxA (7.9%).

6.2. Open pollinated progeny evaluation

First round of testapping of 215 OP seedlings collected in 2017 from the further evaluation trials FETs 2003 and 2005, which comprised preliminary selections from the wild germplasm, interspersed with high

yielding Wickham control clones, revealed seven potential ones, to be confirmed in the second round next year.

6.3. Phenotyping of mapping population for QTL identification

Vacancy filling was carried out in the interspecific mapping population planted in a field trial last year, laid out in a lattice design with four replications for phenotyping for yield and other traits.

7. Other studies

7.1. Studies on alternative sources of natural rubber yielding plants

7.1.1. Ceara rubber (Manihot glaziovii)

Seven germplasm accessions from Palakkad region and four plants multiplied through stem cutting collected from Vaikom region are being conserved at RRII.

ADVANCED CENTRE FOR MOLECULAR BIOLOGY AND BIOTECHNOLOGY (ACMBB)

The Advanced Centre for Molecular Biology and Biotechnology (ACMBB) set up during the XIth Plan period is a functional grouping of scientists working in the areas of Molecular Biology, Biotechnology, Genome Analysis, Molecular Physiology and Molecular Pathology. This was mainly done to functionally merge different labs working in similar areas for better efficiency and saving of resources. ACMBB together constitutes about 15 per cent of RRII research. The ACMBB conducts research on various projects which includes studies on the molecular basis of genetic improvement of natural rubber trees and biotechnological interventions for developing Genetically Modified (GM) rubber. Studies at ACMBB would help to speed up crop improvement, aiding in developing new high yielding, climate resilient and disease tolerant clones faster.

I. BIOTECHNOLOGY DIVISION

The prime objective of research in the Biotechnology Division is attaining crop improvement in *Hevea* expeditiously through biotechnological interventions. Genetic transformation protocols were developed in rubber and fine-tuned for developing *Hevea* transgenics integrated with agronomically important genes. Genetic improvement by the incorporation of desirable genes for imparting enhanced biotic and abiotic stress tolerance, improving latex yield and growth has been accomplished. Perfecting the system for development of antibiotic marker free transgenics was also attempted. Research programmes aimed at developing *in vitro* techniques to complement conventional breeding were also executed. Other than these, development of protocols for the propagation of new elite *Hevea* clones *via* somatic embryogenesis from different explants, development of ploidy variants, *in vitro* approaches for disease tolerance and cloning and characterization of genes are also being envisaged.

1. Development of Transgenic Plants

1.1. Genetic transformation of *Hevea brasiliensis* with HbMnSOD gene for stress tolerance

Development of transgenic plants tolerant to abiotic stresses was attempted through genetic transformation using the embryogenic callus of clone RRII 430 as the initial explant. Six transgenic cell lines were emerged from the infected calli and GUS assay was performed in the developed callus. All the transgenic cell lines were GUS positive. Putatively transgenic cell lines were multiplied in the proliferation medium containing the selection antibiotic Kanamycin (350 mg l^{-1}).

Permission was obtained from the Genetic Engineering Approval Committee (GEAC), Govt. of India to conduct confined field trials of the MnSOD transgenic plants (clone RRII 105) at the Regional Research Station, Guwahati, Assam. The flanking sequences of the MnSOD transgene in the developed transgenic plants were determined.

1.2. Genetic modification with *hmgr*1 gene for improved latex yield

Agrobacterium mediated transformation using the embryogenic callus of clone RRII 430 and RRII 105 produced transgenic cell lines at a frequency of 20 per cent. The cell lines were subcultured to the proliferation medium, but failed to multiply. New transformation experiments were repeated with embryogenic callus of clone RRII 430 and overgrowth free callus was maintained in the selection medium for the emergence of new transgenic cell lines.

1.3. Genetic transformation of *Hevea brasiliensis* with osmotin gene

Molecular analysis for gene integration of already developed transgenic plants was completed. In order to distinguish the native osmotin gene from the transgene, experiments were done to amplify the native osmotin gene. Primers were designed based on the available literature, and native osmotin gene was amplified from the genomic DNA of the clone RRII 105. Two alleles of the gene were amplified from RRII 105 and sequenced. On sequence analysis, it was proved that native gene is intronless and both the alleles are distinct from the inserted osmotin gene. Thus the uniqueness of the inserted osmotin gene was proved. The sequences were deposited in the Gen Bank. Attempts are being made for studying the expression of transgenes.

Experiments are continuing for developing transgenic plant integrated with osmotin gene from clonal material. *Agrobacterium* infections were carried out using anther derived embryogenic calli and cultures were maintained in selection medium. Embryos were induced from already developed transgenic lines. Application for conducting a mini field trial with already developed transgenic plants were drafted.

1.4. Development of *Hevea* transgenics with IPT gene for TPD tolerance

Agrobacterium mediated transformation experiments with iso-pentenyl transferase (ipt) gene for enhanced vegetative growth as well as TPD tolerance were continued. Actively proliferating embryogenic callus derived from leaf explants of clone RRII 105 and RRII 414 was infected with the bacterial culture. Cysteine (100 mg l^{-1}) and phyto hormones were included in the induction medium. After co-cultivation infected tissues were washed thoroughly with sterile water to prevent bacterial overgrowth. New transgenic lines were obtained with good frequency (15%). Proliferation of transgenic

lines was carried out in selection medium by reducing the con of 2, 4-D from 2.0 to 0.8 mg l^{-1} and increasing the sucrose level from 20 to 50 g l^{-1} and phytagel from 2.5 g l^{-1} to 4.0 g l^{-1}. Embryogenic callus initiation and further embryo induction were obtained in standardized medium with minor modifications. All experiments for transgenic callus proliferation and embryo induction were carried out in the presence of Kanamycin (200 mg l^{-1}). Embryo induction from the transgenic callus was obtained with a frequency above 50 per cent. After embryo induction, the concentration of Kanamycin was reduced to 100 mg l^{-1} for embryo maturation and plant regeneration. Attempts are being made for successful hardening of the transgenics.

1.5. Development of biotic/abiotic stress tolerant plants with Hsp31 gene

Experiments were continued to regenerate transgenic *Hevea* plants incorporated with Hsp31 gene for imparting abiotic/biotic stress tolerance. Embryogenic callus of clone RRII 105 and RRII 414 obtained from leaf cultures were used as target tissues. Callus to be used as target tissues for *Agrobacterium* infection were proliferated by subculture in fresh medium supplemented with stress inducing compounds such as poly ethylene glycol and enhanced level of phytagel (4.0 g l^{-1}). On the day of bacterial infection the callus was transferred to petri plates and desiccated for one hour by slow drying in the laminar flow hood before *Agrobacterium* infection. The infection medium was supplemented with picolinic acid and phytohormones such as BA, 2,4-D, NAA and IAA. The infected tissues were thoroughly washed with sterile water for controlling bacterial overgrowth. Regular subculture of infected calli in fresh selection medium were carried out and putatively transgenic lines were emerged from the infected tissues with a high frequency (20%). All the transgenic lines obtained were proliferated by regular subculture. Embryo induction was attempted from the proliferated callus by supplementing phytohormones, PEG and increased level of agar. Plant regeneration could be obtained *in vitro* from the transgenic embryos with a frequency of around 5 per cent. Different methods are being tried for successful hardening of the transgenic plants.

1.6. Genetic transformation of *Hevea* for enhanced biotic and abiotic stresses by the manipulation of epicuticular wax through Shine (SHN1) integration

As a preliminary step, epicuticular wax was quantified from different developmental stages of leaves from *Hevea benthamiana* and RRII 105 in two seasons (winter & rainy) from two different sources (field grown trees and budded plants) and no significant variation in wax content among *benthamiana* and RRII 105 was noticed. Work will be repeated. Simultaneously, to examine the role of epicuticular wax on drought and disease resistance, work was initiated. To relate any difference at the molecular level, work was also initiated from few clones having high and low wax content.

1.7. Genetic transformation with S-6-PDH for abiotic stress tolerance

Inoculation of immature anther was carried out for clone RRII 430. Callus was induced and cultured for proliferation. Overnight inoculum of **S-6-PDH** gene was prepared and proliferated callus of clone RRII 430 was infected, co-cultivated and transferred for selection. Putatively transformed cell lines obtained were transferred for proliferation and embryogenic callus .

1.8. Development of marker free transgenics in *Hevea brasiliensis*

Attempts for developing the gene constructs with marker free system could not give any positive results. Hence, attempts to verify the sequence fidelity of the plasmids were made. Plasmids brought from Arkansas University were extracted and used for *E. coli* transformation. Positive colonies were identified by colony PCR and glycerol stocks were prepared. Plasmids were isolated from this culture using standard protocol and send for sequencing. After sequencing and identification of the restriction enzyme sites, it was found that enzymes sites in the multiple cloning site except *mlu1 was* present in the vector backbone. Hence, mlu enzyme was selected for developing new constructs.

Fresh plasmids were isolated from *E.coli* using standard procedure. Plasmids were mobilized in to *Agrobacterium* by freeze thaw method. Positive colonies were selected by colony PCR and glycerol stocks were prepared. New transformations experiments were performed for the validation of the construct in *Hevea* callus.

1.9. Gene identification for development of climate resilient transgenic plants

The plasmid vector (pEGAD) containing translationally controlled tumor protein (TCTP) gene was developed by Dr. P. Venkatachalam and supplied through Material Transfer Agreement (MTA). E. coli competent cells were prepared and transformed with the plasmid vector pEGAD. Plasmid isolation was carried out from the recombinant colonies using alkaline lysis method described by Birnboim and Doly (1979). PCR analysis was performed to amplify the TCTP gene using gene specific primers designed based on the reported cDNA sequence information in NCBI (Accession number DQ 323740). Competent cells of *Agrobacterium* strains EHA 105 and EHA 101 were prepared and used for transformation by freeze thaw method. Recombinant colonies were screened for the presence of inserted TCTP gene by colony PCR using gene specific primers. Positive colonies were grown in semi-solid LB medium and stored as glycerol stock at -80°C.

1.9.1. Restriction enzyme digestion

Recombinant plasmid (1 µg) was digested with 1µl restriction enzyme, EcoR1 (10,000 U) with appropriate buffer and BSA. Restriction digestion was continued overnight at 37°C and the enzyme was inactivated. The fragments were size fractionated on a 0.8 per cent agarose gel containing 0.1 per cent (w/v) ethidium bromide and viewed under a uv transilluminator and documented. Digestion with single enzyme released the inserted TCTP gene of length 670 bp. Results indicated that the gene of interest was inserted at the EcoR1 site in the multiple cloning site (MCS) region of the plasmid.

1.9.2. Sequencing of the recombinant plasmid

Plasmid vector pEGAD containing the insert was diluted to a concentration of 150 ng/µl and sequenced using gene specific primers. The chromatogram was analysed using the DNA baser assembler tool and the data was examined using the forward and reverse primers. Restriction sites for EcoR1 enzyme was identified at the left and right border. The complete sequence of the TCTP gene was identified between the Eco R1 restriction sites.

1.9.3. Determining the optimum level of glufosinate for selection

Selection antibiotic glufosinate ranging from 20 mg l^{-1} to 120 mg l^{-1} were included in the culture medium. Control

callus (clone RRII 430) was sub cultured in different media combinations and the concentration of glufosinate inhibiting the growth of control calli was identified. Experimental observations showed that glufosinate (100 mg l^{-1}) in the culture medium suppressed growth of the callus and therefore chosen as the optimum concentration for selection of transformants (Fig. Biotech. 1).

1.10. Gene editing

A new project on CRISPR Cas mediated gene editing was initiated. A project proposal on CRISPR Cas 9 mediated gene editing was sent to DST for external funding.

2. Propagation of elite *Hevea* clones

2.1. Somatic embryogenesis from immature anther of RRII 400 series clones

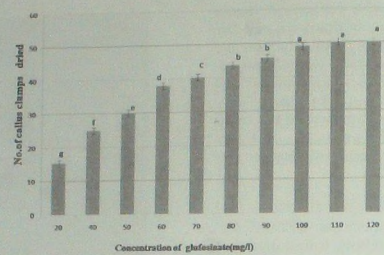
During the reporting year, work was imitated to develop plants from 400 series clones. Immature anther collected from *Hevea* clones RRII 414, 417, 422 and 430 were inoculated on callus induction medium containing 2,4-D (2.0 mg l^{-1}) and BA (2.0 mg l^{-1}). After 40-50 days of culture, induced callus from all clones was cultured for proliferation on the same fresh medium and got proliferated. Proliferated callus were subcultured for embryogenic calli formation / embryogenesis.

Preliminary experiments carried out during last year with coconut water (CW) and calcium were repeated in this current year to confirm the results. To investigate the effect of CW, immature anther from *Hevea* clone RRII 414 were inoculated on callus induction medium containing 0, 5%, 10% CW. Fifty anthers were inoculated on each combination and the experiment was replicated 2 times. After 40 days, cultures were evaluated. Results indicated that callus was also induced without CW. The callus induction frequency, growth and proliferation of callus was almost similar to that of medium containing 5% CW. However, a slight enhancement on the response was noticed with 10%.

M: Marker ; 1-2: Uncut plasmid;
3: Plasmid digested with EcoR1

Fig. Biotech. 1. Restriction digestion of plasmid pEGAD with Eco R1

Experiment with calcium (nitrate and chloride forms) was also repeated by supplementing calcium nitrate and chloride in three combinations with 3, 6, and 9mM concentrations. Immature anther from RRII 414 was inoculated in these combinations and experiment was repeated twice. Cultures were kept for callus induction for 60 days under darkness. Results of the study showed that callus induction frequency, growth and proliferation was similar in all three combinations in both forms of calcium. However, the texture of callus was different. Compared with 3mM, 6 and 9mM produced



The vertical bars with the common letters are not significantly different at p<0.05,Means are calculated using DMRT;Vertical line indicates the error bar

Fig. Biotech. 2. Effect of glufosinate on growth of *Hevea* callus

highly friable and loose callus. Both forms of calcium showed more or less similar response indicating that enriching the callus induction medium with either CaNO3 or CaCl2 in 6mM-9mM was good for friable callus induction.

2.1.1. Encapsulation of somatic embryos and synthetic seed production

Somatic seeds are nothing but encapsulated somatic embryos. Hence a large number of somatic embryos were needed. For inducing somatic embryos, work was initiated from RRII 105. Immature anther from RRII 105 were collected, inoculated on callus induction medium containing 2.0 mg/l 2,4-D and 0.5 mg/l kn and kept the cultures under darkness. After 50 days of culture, callus was induced. Induced callus was proliferated on the same fresh medium and then subcultured for 4 months for embryogenic callus formation on medium with 0.3 mg/l kin, 0.3 mg/l BA and 0.3 mg/l NAA . Embryogenic callus induced were further transferred to embryo induction medium and somatic embryo induction awaited.

2.2. Somatic embryogenesis from leaf explants

In vitro leaf cultures initiated with clone RRII 105, RRII 414 and RRII 417 collected from glasshouse grown bud grafted plants could induce callus after 3-4 weeks in earlier standardized callus induction medium. Rate and time of callus induction was found to vary with the clone. Callus induction with good frequency (80%) was obtained in clones RRII 105 and RRII 417. In another experiment, explants were initially cultured in callus induction medium supplemented with high auxin (2,4-D 10 mg/l) concentration for two weeks and then sub cultured in callus induction medium with earlier standardized concentration of auxin (2,4-D 1.5 mg/l). Here the explants swelled with nodules and callus induction occurred within three weeks in all clones. However, RRII 417 gave maximum response with 100 per cent callus induction

within three weeks and also increased callus formation. Proliferation of the callus was attempted as carried out in earlier experiments through repeated subculture in medium with gradual increase in cyotokinin\auxin ratio and sucrose concentration. Towards the end of the subculture period for callus proliferation, stress inducing compounds such as poly ethylene glycol (5.0 g/l) and phytagel (3-4 g/l) were also supplemented in the medium. Since in earlier experiments, addition of adenine hemisufate at a concentration of 50 mg/l in the callus induction medium, 100 mg/l in the callus proliferation medium and 185 mg/l in the embryo induction medium favored embryogenesis, this was also supplemented. Embryogenic callus induction and somatic embryogenesis were attempted in the earlier standardized medium for clone RRII 105. For clones RRII 414 and RRII 417, minor modifications were made. The amino acids asparagine (150 mg/l) and glutamine (800 mg/l) which were found to enhance the frequency of embryo induction in earlier experiments were also included in the medium. Usually induction of embryos was observed with a good frequency, on the medium surface, one month after culture when the medium started desiccation. The embryos when transferred to modified WPM medium supplemented with folic acid (0.5 mg/l), organic supplements and phyto hormones (BA, GA_3 and IBA) got enlarged and became cotyledonary after two weeks. These somatic embryos were then subjected to slow desiccation by keeping them in sealed sterile petri plates for two days. On transfer of the desiccated embryos back to the earlier medium, they germinated with bipolar differentiation. *In vitro* plant regeneration was obtained in MS medium containing BA, GA3 and IBA and acclimatization of the plants is being attempted in soilrite under humidity controlled conditions.

2.2.1. *Effect of salicylic acid on somatic embryogenesis*

The experiment to study the effect of salicylic acid on somatic embryogenesis was repeated. Pretreatment of the explants with salicylic acid as well as addition in the callus and embryo induction medium was experimented. It was observed that callus induction with improved texture was obtained in 20% of the cultures. Since salicylic acid at a concentration of 2.0 mg/l favored formation of proembryo like protrubances in earlier experiments, the effect of this compound in presence of different phytohormone concentration was also experimented. The explants were initially exposed to higher concentration of 2,4-D (2-10 mg/l) and BA (2- 5 mg/l) in presence of salicylic acid (2.0 mg/l) both as a pretreatment for 3-5 minutes as well as by addition in callus induction medium and culture incubation for 20 days. The callus obtained was proliferated as in earlier experiments with addition of 1.0 mg/l salicylic acid in the proliferation medium along with the phytohormones BA, 2,4-D and NAA. As in earlier experiments, addition of 2.0 mg/l salicylic acid in the callus induction medium favored explant swelling and callus induction with improved texture. Presence of salicylic acid during callus proliferation also aided embryogenic callus initiation. Experimentation on proembryo induction in leaf cultures in presence of salicylic acid is being continued.

2.3. *Ex vitro* adventitious rooting for production of self rooted clones

Bud grafted plants were produced from somatic plants. *Ex vitro* rooting of shoots collected from these plants maintained in polybags in the glasshouse were attempted.

Different combination and concentration of phytohormones such as BA, TDZ, IBA and NAA were given as hormone pulse for different time intervals. The synergistic effect of fresh coconut water on root induction was also experimented. Experiments were continued to identify suitable conditions and potting medium. Sterile soilrite was found to be more suitable for root induction when hormone treated shoots were maintained in these under humidity control. Both BA and TDZ favoured adventitious rooting in presence of IBA and NAA. Effect of Paclobutrazol on root proliferation after root induction is being attempted.

3. Induction of ploidy variation in *Hevea brasiliensis* through *in vitro* techniques

3.1. Development of homozygous diploids in *Hevea*

3.1.1. *Embryo sac culture for the development of gynogenic haploids*

Isolation and culture of embryo sacs from mature female flowers of different clones of *Hevea brasiliensis* was carried out with the objective of developing haploids/ homozygous diploids. Callus induction could be obtained in all the clones tried, though the frequency of callus induction as well as the texture of the calli varied among the clones. Embryos could be regenerated from three clones RRII 105, 414 and PB 330. Plant regeneration was obtained for clones RRII 105 and 414. These plants were subjected to acclimatization. Haploid nature of the plants was confirmed through cytological analysis. Homozygous diploids which are ideal candidates for elite clone breeding in *Hevea*, can be developed from these haploids through chromosome doubling. Gynogenic haploids with confirmed ploidy (n=18) have been regenerated through embryo sac culture for *Hevea* clones RRII 414 and RRII 105, for the first time. Haploid plants of clone RRII 414 have been successfully established in the field.

The culture of isolated embryo sac of clone RRII 105 in the callus induction medium after a short exposure to colchicine resulted in the development of friable callus from the embryo sac cells. The ploidy of the callus was ascertained by flow cytommetry and confirmed as mixaploids. Proembryogenic masses developing from the callus were sub cultured in the embryo induction medium.

The haploid callus of clone RRII 422 produced somatic embryos in the embryo induction medium and matured embryos with bipolar differentiation in the germination medium.

4. *In vitro* approaches to complement conventional breeding programmes

4.1. Induction of Polyembryony

4.1.1. *Development of uniform seedlings*

Immature fruits were inoculated during the current season for inducing multiple embryos/embryogenic callus. Experiments were continued for developing uniform seedlings of known parentage through *in vitro* pollination. Inflorescence were collected from field grown trees and the cut end was dipped in distilled water. Stigmatic pollination was carried out with freshly opened flowers. After 48 hrs, flowers were sterilized and fertilized ovules were isolated using standard protocol and inoculated in the media. Calli obtained were subcultured for embryo induction.

Yield recording in the polyembryony derived plants is continuing. The second year yield data recording was completed in the field planted polyembryony derived seedlings. Poly plants had lowest CV among all treatments indicating their better uniformity.

4.1.2. Stock Scion interaction studies

Leaf samples were collected from RRII 105 plants of different age groups were collected from the same geographical area and subjected to epigenetic analysis. Polymorphism could be identified in the epigenetic profile among the trees.

5. *In vitro* screening of rubber clones for disease tolerance

5.1. *In vitro* approaches to impart *Corynespora* leaf fall disease (CLFD) tolerance

5.1.1. *In vitro selection of Corynespora tolerant lines of clones RRII 105 and RRII 414 through toxin challenge*

For the first time, CLFD tolerant plants were regenerated through somatic embryogenesis from cassiicolin toxin habituated callus in an otherwise susceptible *Hevea* clone RRII 105. Embryogenic calli of clone RRII 105, the most popular and widely cultivated clone, which at the same time susceptible to Corynespora leaf fall disease (CLFD), were cultured over medium fortified with different levels of cassiicolin toxin. Surviving calli obtained after two cycles of selection were transferred to toxin free media. Plants could be regenerated from these calli and were acclimatized and transferred to soil. Laboratory level bioassays confirmed improved tolerance of the regenerated plants towards CLFD. Regenerated plants were successfully established in the field. Multiplication through bud grafting has been achieved and the bud grafted plants also exhibited improved tolerance towards CLFD in the lab level bio assay.

6. *In vitro* selection and development of drought tolerant plants

In vitro culture of tissues are an ideal tool for *in vitro* screening and selection of desirable characters. This was achieved by selection agents such as PEG and the tolerant lines were screened, selected and regenerated plants. For producing drought tolerant, high yielding RRII 414 clone, two experiments were carried out. In the first experiment, immature anther from RRII 414 were inoculated in standard callus induction medium and kept cultures for callus induction. Induced callus were proliferated and approx. 500 mg callus was subcultured in medium containing PEG in four combinations (0%, 5%, 7.5% and 10%) and results awaited.

In the second experiment, immature anther from RRII 414 were directly inoculated in callus induction medium with different levels of agar (5, 6 & 7 g l^{-1}). Cultures were kept for callus induction under darkness for 60 days. In each treatment, 5 explant / tube was inoculated and for one combination 50 explants were inoculated and experiment was repeated 2 times. Cultures were kept for callus induction . With 0.5% agar, little callus was induced and were cultured for proliferation. However, with 6 and 0.7 g l^{-1} agar no callus was induced.

II. GENOME ANALYSIS LABORATORY

Ongoing research projects in the Genome Analysis Laboratory are grouped under four major areas *viz.*, (1) development, optimization and validation of molecular tools for the assessment of genetic diversity and evolutionary relationships in rubber and genome mapping (2) development of genetic markers for biotic and abiotic stress tolerance and understanding the stress adaptation processes through transcriptome analysis (3) cloning and characterization of agronomically important genes and (4) rubber genome sequencing and *de-novo* assembly. Besides the above research programs, a collaborative project has been initiated with CSIR-NEERI on conversion of tropical forests to rubber plantations in Kerala and its impact on the soil environment and different eco-restoration strategies.

1. Development, optimization and validation of molecular tools for the assessment of genetic diversity in rubber, clonal identification and genome mapping

1.1. Single nucleotide polymorphisms (SNPs) in *Hevea*

1.1.1. SNP identification and haplotype structuring in the latex biosynthesis genes of Hevea brasiliensis

Characterization of the highly distorted *PMVK* gene indel locus reported last year was continued. In order to establish the role of male and female parents in the distorted segregation of PMVK gene alleles, a progeny population derived by crossing PB 280 x RRII 105 was subjected to segregation analysis. Genomic DNA from 23 progenies was isolated and genotyping was done using the PMVK indel marker followed by segregation pattern analysis. Contrary to the skewed segregation ratio obtained earlier in the other two populations, the segregation ratio of alleles in this progeny population agrees well with expected Mendelian ratio. To further confirm the maternal effect of RRII 105 in the distorted segregation of PMVK gene alleles in its progeny populations, another population of parents RRIC 100 and IRCA 18 having the same allele combination as that of the previous populations was genotyped. Since the female parent RRIC 100 have the same allelic combination as that of RRII 105, the segregation pattern in this progeny population will prove whether the segregation distortion is specific to the female parent RRII 105 or not. DNA was isolated from 34 progeny plants and genotyping using the PMVK indel marker is in progress.

Validation of putative SNP markers for yield:

Putative SNP markers associated with higher expression levels of genes from the mevalonate pathway were reported in previous years. In order to use them as potential markers for yield, they have to be validated in a set of known high yielding and low yielding genotypes. A total of 14 genotypes consisting of high yielding clones and consistently low yielding wild accession were selected for the preliminary analysis. Leaf samples were collected from these genotypes and genomic DNA was extracted from the leaf samples, quantified and stored for HRM SNP genotyping.

1.1.2. Development of a rapid clone identification system based on SNP markers

Proper identification of clones plays a vital role in crop management systems and

research in natural rubber (*Hevea*). However, the traditional approach of clone identification based on morphological differences poses several limitations, as most of the morphological characters are influenced by the environmental conditions and the age of plant, and they are not variable enough to adequately characterize genetic differences among elite genotypes. Moreover, due to the narrow genetic base of present breeding population, the popular cultivated clones of rubber does not exhibit highly conspicuous and very distinct variations in its characters making it difficult to identify clones at morphological level accurately. Though molecular markers can clearly differentiate the genetic material avoiding any of the environmental influences, the current marker technologies in rubber required lengthy laboratory procedures which may takes several days. In this context, a rapid clone identification based on SNP information from popular clones assumes relevance. In order to identify clone specific SNPs in *Hevea*, selected highly polymorphic regions of two important rubber biosynthesis genes with maximum heterozygosity, was re-amplified from 15 most popular rubber clones. PCR products were purified, quantified and sequenced. Sequence analysis is in progress.

1.2. Genetic authentication of clone museum using molecular genetic markers

The 'Wickham' germplasm collection comprising around 180 clones in the Clone Museum at RRII, has been serving as a source of experimental material (budwood, leaf tissue, twigs) of different clones – both obsolete and elite, for different Divisions in the Institute over the years. Each accession had been planted in a row with 10 points each. In view of the various experiments that the materials are being used for, genetic authentication of the identity of each plant point is of utmost importance. The narrow genetic base and high within-plant variation in leaf morphology make a definite identification very difficult. Therefore the laboratory initiated the validation of these germplasm materials using molecular markers. Genomic DNA was isolated from more than 125 plants belonging to more than 55 genotypes. The quality of the DNA samples was checked, diluted and kept ready for marker studies.

RAPD reactions were set using 10 different primer combinations to estimate the genetic uniformity of plants belonging to genotypes like Tjir 1, Mil 3/2, PB 86 and RRII 33 along with control DNA samples. Analysis of the banding pattern indicated mixing-up of clones in Tjir 1 set whereas all the plants of Mil 3/2 appeared to be true to type. All the plants of PB 86 and RRII 33 appeared to be true to type. DNA isolation and marker analysis of rest of the samples is in progress.

2. Characterization of stress-tolerant clones of *Hevea* using molecular markers and understanding gene regulation under abiotic stresses

2.1. Methylation dynamics of *Hevea brasiliensis* genome

2.1.1. *Identification of epigenetic markers for abiotic stress in Hevea*

In order to identify epigenetic changes accumulated over a period of 30 years within the genome of clone RRII 105, DNA methylation profiling of RRII 105 plants planted during the 80s, 90s and after 2010 was attempted using MSAP technique. Genomic DNA was isolated from mature trees planted in the same region during the aforementioned periods. DNA was subjected to double digestion with *EcoRI*/

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MspI & *EcoRI/HpaII* and ligated with respective adaptors. Pre-amplification was carried out using selective primers for further analysis. Selective amplification using a total of 14 different primer combinations were performed on the DNA from nine RRII 105 plants planted at different time intervals. The samples were run on denaturing PAGE gel and documented. Clear evidence for the presence of DNA methylation polymorphism was observed among the analyzed plants. Specific year-wise patterns were detected in one primer combination. The bands showing consistent DNA methylation polymorphisms (epialleles) obtained by the MSAP analysis were eluted from acrylamide gel. Re-amplification was performed using respective primer combinations and the re-amplified fragments were purified and cloned for plasmid isolation. Plasmids with the fragments showing consistent DNA methylation polymorphisms (epialleles) were isolated from multiple positive colonies and kept ready for sequencing.

2.3. Functional genomic studies in *Hevea*

2.3.1. *Identification and validation of fungal transcripts obtained from Hevea root transcriptome data*

AM fungi receive increasing attention for their potential use in sustainable agriculture because they interact with most crop plants to increase crop yield and nutrient uptake, enhance resistance to plant pathogens, and stabilize soil structure. Therefore identification of beneficial AM fungal species associated with *Hevea* root tissues and rhizosphere is very important. In order to verify their association with plant age, attempts were made to detect their presence in root of rubber plants of different age (5 months to 5 years).

Fourteen primers based on fungal ribosomal and ITS region were synthesized to allow species level resolution of AM fungal communities within root tissues of *Hevea*. Four primer mixtures were prepared by mixing them in various combinations. The primer mixtures were used to amplify a barcoding region based on ribosomal and ITS region of AM fungi from the DNA extracted from root tissue of two sets of rubber plants (one year old seedlings & 5 year old trees). Expected amplicon of 1.8 kb was obtained from all the samples, which were purified and cloned in pGEMT vector. 92 plasmid samples harbouring partial ribosomal and ITS region were isolated and sequenced to establish the species level diversity of AMF in root tissues of *Hevea*.

Phylogenetic analysis using softwares like ClustalW and Phylogeny was attempted. After the final analysis, we could identify a total of 14 different species of AM fungi from the root tissues of *Hevea* plants of age 1 year and 5 years. The different species identified mainly comes under the families *viz*. *Glomeraceae*, *Gigasporaceae* and *Aculosporaceae*. The presence of one species belonging to the comparatively rare *Paraglomeraceae* family was also identified. We also noticed the presence of 7 uncluturable AM fungal species within the root tissues of *Hevea*. Cluster analysis revealed that except one specie, all grouped under any of the above four families. Species wise sub-clusters specific to plant age was observed during sample source based phylogenetic analysis, but further confirmatory tests are required to establish their association with age of the plants. Sequence of approximately 1.35 kb length

of good quality from the ribosomal and ITS region of AM fungal species identified from the root tissues of *Hevea* were edited and kept ready for submission to genbank.

3. Cloning and characterization of agronomically important genes

3.1. Cloning and characterization of *WIN1/SHN1* gene from *Hevea*

INDUCER1/SHINE1 (WIN1/SHN1) is a transcription factor of the ethylene response factor (ERF) family. This transcription factor is known to trigger epicuticular wax production thereby enhancing drought tolerance in several plants. Attempts were made to characterize two isoforms of *WIN* genes (*WIN1* and *WIN1X*) from the genomic DNA of clone RRII 105 and *H.benthamiana* last year In order to verify whether there is any association between sequence polymorphism identified in two isoforms of *WIN* genes (*WIN1* and *WIN1X*) from Hevea genotypes having differen wax content, attempts were made to amplify them from two more genotypes (RRII 33 and *H. Spruciana)*. The amplified products were cloned and plasmid was extracted and purified for sequencing.

4. Metagenomics and microbe identification in rubber ecosystems (Collaborative project with CSIR-NEERI)

The fungal ITS region was successfully amplified from all the 130 samples from five different land types (Weeded rubber, un-weeded rubber, rubber with cover crop, cocoa rubber intercrop and forest) using fungal ITS region specific primers. Raw sequence data derived from these samples was subjected to bioinformatics analysis to derive useful information. Preliminary sequence analysis results indicate that microbial concentration is more in the soil collected from rubber plantations having cocoa as intercrop. More than 37 per cent of the fungal OTUs were derived from this sample type whereas the other soil types had 10 to 20 percentage of OTUs. Roughly 50 per cent of the fungal OTUs in all the five soil types belonged to the family Ascomycota. Interestingly more OTUs under beneficial AM fungal family was observed in the cocoa intercrop plantation. As expected, forest soils had more number of unclassified fungal species. Non metric Multidimensional Scaling (NMDS) analysis revealed that forest soil holds more fungal diversity followed by rubber plantation. Though cocoa rubber intercrop plantation is richer in fungal population, they seems to hold less diversity when compared to other land types. Further analysis to characterise them is in progress.

5. Development of genetically modified rubber plants with agronomically desirable traits

5.1. Molecular characterisation of transgenic lines developed

5.1.1. Estimating the site of genomic integration of MnSOD gene in the transgenic MnSOD plants developed by RRII

Detailed molecular characteristics of flanking sequences of insertions play an important role in the safety assessment of genetically modified crops. It is also essential to understand whether native genes are disrupted during the insertion process and to predict and assess its impact on the plant development and function.

The site of integration of the introduced MnSOD gene along with promoter, reporter and antibiotic marker gene in one line of transgenic rubber plant developed (L1) was estimated by genome walking technique. Analysis of the sequenced region and sequence homology analysis with available whole genome sequence information revealed that the introduced DNA fragment got integrated in to the intronic region of a variant of Leaf rust 10 disease-resistance receptor-like protein kinase gene. Preliminary analysis suggest that there exists only a single copy of the inserted fragment in the transgenic *Hevea* genome which corroborates our earlier southern analysis results. In order to further confirm the site of integration of MnSOD gene construct in the transgenic plant, specific primers from the upstream and downstream region of the insert were designed using the whole genome sequence information available in public domain. Sequencing of this border regions confirmed our earlier findings. Confirmatory PC tests were also carried out to test whether the two lines developed (L1 & L2) were originally evolved from two different events or not. Confirmatory PCR tests followed by sequencing of border sequences from both the lines revealed that the site of integration of the transgenes is exactly the same for L1 and L2. These results proved beyond doubt that the lines L1 and L2 originally occurred from a single event and have the same genetic constitution.

5.2. Validation of the Cre-loxP construct for developing marker free transgenic Hevea

The entire region between left boarder and right border of the Cre-loxP construct (PNS14) brought from Arkansas was verified by primer walking. Several differences from the sequence received earlier were detected. All the genes in the construct was found to be intact. The restriction enzymes included in the MCS were mapped in the entire region including the vector backbone sequence. Sequence analysis revealed that except *Mlu1*, all the other five REs in the MCS have multiple recognition sites either in the insert or in the backbone and therefore could not be used for new gene insertion directly to the MCS.

5.3. Molecular characterization of *in vitro* plants developed from toxin habituated callus cultures

Leaf samples were collected and genomic DNA was extracted from two plants developed from toxin habituated callus cultures. In order to confirm their clonal identify (RRII 105), SSR analysis using seventeen different microsatellite primers were performed. The PCR products were analyzed by running it in denaturing PAGE. SSR marker profiles of the two samples with all the marker loci clearly revealed that the tested samples were very similar to the reference sample *i.e.*, the clone RRII 105.

PLANT PATHOLOGY DIVISION

The Division is mainly concentrating on studies on economic and eco-friendly management of pests and diseases. Evaluation of new clones for disease resistance, identification of genes and QTLS for disease tolerance, role of biotic etiology of tapping panel dryness. Development of management practices on newly emerging diseases. Experiments on crown budding technique by which the crown of a high yielding clone is modified by developing a canopy with high disease tolerance. Yield loss studies due to major leaf diseases. In addition to research, the division also takes up testing of spraying equipment, plant protection chemicals and analysing water samples for estimating bacterial population. Training on disease management, maintenance of spray equipment and apiculture are the other activities undertaken by the Division.

Advisory work on disease management is also undertaken through field visits, telephonic advisory, WhatsApp and Online Rubber Clinic. About 1350 cases were attended through WhatsApp and 104 water samples were analysed during the reporting year.

1. Leaf diseases

1.1. Abnormal leaf fall disease

1.1.1. Disease survey

Abnormal leaf fall survey was carried out during 2020 disease season in Kerala and South Karnataka. During the survey, it was observed that low leaf fall incidence was recorded in all the clones and regions. No difference was observed between the clone RRII 105, RRII 414 and RRII 430. The combined infection of CCLS and ALF was observed in many plantations across the traditional area during monsoon season.

The impact of abnormal leaf fall on growth and yield of four modern clone's *viz.*RRII 414, RRII 422, RRII 429 and PB 260 gave varying results. The severity of ALF disease in general during 2020 was low. Among the clones, high leaf fall of 60-90% recorded in unprotected blocks of RRII 414, RRII 422, RRII 429 and PB 260.The yield of the trees in the sprayed blocks continued to be significantly higher in clones of RRII 429, RRII 414 and PB 260.

The yield variation was noticed among the clones due to ALF at CES Chethackal. The ALF was more in the clone RRII 429.The crown budded trees in CES Chethackal recorded higher yield than control PB 260.The yield loss data of the clones are presented in Table Path. 1.

Table Path. 1. **Yield loss due to abnormal leaf fall, CES Chethackal (2020-21)**

Sl. No.	Clone	Yield loss (%)(2020-21)
1	RRII 414	24.4
2	RRII 429	30.0
3	RRII 422	17.2
4	PB 260 (Control)	26.5
5	Crown bud	39.7

1.2. Integrated control

The endophytic bacteria (RH 34) screened against *Phytophthora* was tested for its intrinsic tolerance to COC and the agricultural spray oil. The RH 34 could establish in both. The same was prepared in talc formulation. The half-dose of the recommended oil-based COC and spray oil was mixed with the bio-agent talc formulation and sprayed prophylactically,

CES during 2020. The leaf retention was comparable to the recommended dose. The effect will be evaluated in the multilocational trial for further confirmation.

1.3. Field crown budding

Field crown budding was standardized in all modern clones at RRII farm (RRII 105, 414, 417, 422, 429 and 430) with FX 516 crown. Budding was carried out at 8-10 feet height. An average of 70 per cent success obtained in the field crown budding (Fig. Path. 1)

1.4. Corynespora disease

Nursery evaluation of bioagents (endophytic bacteria) and integrated control against Corynespora leaf fall disease on clone RRII 105 was carried out at Ulickal nursery. The endophytic bacteria were applied as in broth ($1x10^9$/ml) formulation with carbendazim (500 mg). The disease incidence was low during the season.

In vitro and field screening of modern clones RRII 414 showed more tolerance against Corynespora. Leaf infection and leaf fall were noticed in RRII 430 and severe in RRII 422.

1.4.1. Whole genome sequence of virulent Corynespora isolates

Highly virulent Corynespora isolate (from Sheradi) selected for genome analysis. The DNA of this virulent isolate was used for genome sequence (Illumina Hiseq). The genome size of the *Corynespora* isolate is 47.62 MB. The majority of genes detected are part of carbohydrate and aminoacid metabolism. Virulent genes were identified by comparing the predicted proteins with database of fungal virulent factors (DFVF) and found 1026 possible virulent gens.229 of virulent genes having disease key "leaf spot" which is predominant in rubber plantations. The *Cas* gene in the virulent Corynespora isolate is closely related to CC004 (Corynespora isolates) from China.

1.5. New Colletotrichum Circular Leaf Spot

The outbreak of this disease was first noticed during the beginning of July 2017

Fig. Path. 1. Field crown budding and sprouting of crown buds on modern clones

at Poovarani, Paika, Pala, Kerala. The symptoms appear like circular spot of 0.5 to 2 cm diameter. The leaves turn to yellow and fall off. Mostly the lower layer of leaves of both mature and immature plants is affected. The disease was observed in a mild to severe form in Pala, Erattupetta, Poonjar and Ponkunnam regions. Preliminary survey was carried out to understand the spread of the disease. Similar circular spots were observed in some crops. In 2019-20, the disease incidence was observed in the plantations of Thrissur, Idukki, Kottayam, Ernakulam and Pathanamthitta districts and Punalur region. In 2020, the disease was observed in six districts in Kerala State and one district in Tamil Nadu (Figs. Path. 2 A & B).

1.5.1. Management of Colletotrichum circular leaf spot (CCLS) disease

A. *In vitro* evaluation of fungicides

Among the 6 fungicides tested, Thiophanate methyl and Mantram showing 100 per cent growth inhibition even after 10 Day of inoculation followed by Mancozeb, Propineb and COC.

Field evaluation

Fungicide evaluation was done in bud wood nurseries using water base fungicides in the infected bud woods nursery, Pala. Fungicides tested were Indofil, Tagstin and Folicur. All fungicides checked the further development of CCLS

Water based fungicide evaluation in the field was carried out at Mallikassery, Paika using the Clone RII 430 (6 years). The fungicides included were Indofil, Mandiram and Thiophanate methyl. The fungicides were sprayed at 15 days interval using single man carrying mistblower. The result indicated that three fungicides were found to be better than unsprayed control. At Chengalam, Paika the RRII 105 (12 years) were sprayed with two fungicides *viz.* Indofil M-45 and Thiophanate methyl. Two rounds of spraying were undertaken using four men carrying mistblower. These fungicides were found to give better protection.

Combined control of both ALF and CCLS diseases were attempted during 2020

Careful observation in the plantation after summer showers and prophylactic spraying of Oil-based COC were carried out as per our recommendations using mistblowers at the dose of 8 kg Copper Oxychloride in 40 L spray oil/ha. Prophylactic spraying was carried out at three hot spot locations *viz.*, Poovarany (RRII 105 & RRII 414), Chengalam (RRII 105) and Mallikassery (RRII 430) and also at CES (RRII 414, RRII 422, RRII 429, RRII 105 and PB 260). The leaf retention was assessed (Table Path. 2). One prophylactic spraying can control both leaf diseases.

Table Path. 2. **Leaf retention in different locations**

Location/Clone	Leaf retention (%)	
	Sprayed	Unsprayed
CES, Chethackal		
RRII 105	80	10
RRII 414	90	30
RRII 422	75	10
RRII 429	80	10
PB 260	75	20
Poovarani, Pala		
RRII 105	90	30
RRII 414	60	30
Chengalam		
RRII 105	80	40
Mallikasseri, Pala		
RRII 430	80	50
RRII 430 (4 year old trees)	70	20

A B

Fig. Path. 2 A. Symptom on rubber leaves: 2B. Affected plantations

1.6. Powdery mildew disease

1.6.1. Integrated control

The endophytic bacteria were screened for the tolerance to fungicides. We found RH 34 was tolerant to the full dose (0.05%) carbendazim. Initial trial was conducted at RRII using RRII 105 nursery plants during the disease season. The bio-agent was prepared in the talc formulation and dusted alone and in combination with fungicide carbendazim. Three rounds of dusting were done at weekly interval. The combined application was comparable to standard recommendation. But the single application of the bio-agent recorded high disease severity. Field evaluation of the same was carried out during 2020 and 2021. The dusting was carried out using micron duster during the refoliation period. Three rounds of the same were done. During 2020 the disease incidence was low and comparison was not made. The integrated treatment was comparable to sulphur dusting during 2021.

1.7. Crop loss due to combined effect of Abnormal leaf fall and powdery mildew disease (PMD)

Powdery mildew disease of rubber caused by *Oidium heveae* Steinm. causes severe defoliation of young leaves during refoliation after wintering. ALF cause heavy defoliation during monsoon period. The resultant poor canopy and vigour of trees reduce yield. However, the combined crop loss of both ALF and PMD was not studied yet. Hence, evaluation of crop loss due to the effect of ALF was carried out at RRS Padiyoor. All package of practices were same till the time of tapping. After this one block was kept as control (*ie.*, no spraying and dusting). One block both spraying and dusting was undertaken every year. The girth and yield data were recorded and estimated. The clones, RRII 105, PB 5/51, RRIM 600 and PB 235 were used in the study. Some year's crop loss recorded in RRII 105 (44%), RRIM 600 (51%), PB 5/51 (35%) and PB 235 (44%).

1.8. Thread blight disease

A survey was carried out during 2020 in the endemic area of Kothamangalam and Thodupuzha regions to understand the spread, incidence and severity of Thread blight disease. The incidence and severity were 10-50 per cent and 20-30 per cent respectively. Isolation of the fungus was made and identified using ITS specific primers and identified as *Pellicularia filamentosa*.

2. Tapping panel dryness (TPD)

As an observational trial, Tetracycline antibiotic (1000 ppm) were applied on the TPD affected bark (Partial/ Full TPD). Certain trees retained latex .The studies are in progress.

3. QTL Marker development for disease tolerance

3.1. Leaf diseases

3.1.1. Abnormal leaf fall disease

Defence signalling network from transcriptomic data of resistant and susceptible clones after challenge inoculation with *Phytophthora* were identified. Pathogenesis-related (PR) proteins were confirmed to be synthesised and accumulated with time to prevent invasion and establishment of pathogen. PR 2 (â-1,3-glucanase), PR 3 (chitinase) and PR 7 (endoprotease) were found to be released to plant intercellular space. Catalyse degradation of structural components in the cell walls of pathogen was established.

The interplay between salicylic acid (SA) and jasmonic acid (JA) pathways in *Hevea-Phytophthora* interaction was unravelled. A *Phytophthora* resistant rubber clone FX 516 and a susceptible clone RRIM 600 were challenge inoculated with *Phytophthora* spp. and RNA sequencing was carried out using Illumina NextSeq 500 platform. The transcriptome data thus derived from resistant and susceptible clones in both control and pathogen challenged conditions were analysed to investigate the expression of key regulators of SA and JA pathways. It was identified that Non-Expressor of Pathogenesis-Related (NPR1) proteins, a key regulator of SA pathway was up regulated in both resistant and susceptible clones under challenged condition, which indicates its major role during infection. Similarly, to identify the role of JA pathway during infection, an effort was made to analyze expression of the Coronatine Insensitive 1 gene (COI 1) which is reported to be involved in degrading the repressor of JA responsive genes. Up regulation was not observed in *Phytophthora* challenged condition in both resistant and susceptible clones; but down regulation of COI1 was observed in challenged conditions in both these clones, suggesting the potential role of SA in repressing JA pathway during infected condition.

The role of auxin in response to *Phytophthora* infection was also studied as plant hormones play an important role in response to biotic stress. Auxin-signalling was found to act antagonistically towards salicylic acid signalling, which is essential for biotrophic resistance. Up-regulation of auxin responsive factor (ARF) genes in response to *Phytophthora* infection was studied and results indicated that ARF gets upregulated in RRIM 600, the susceptible clone and down regulated in FX 516, the resistant clone during *Phytophthora* infection, a possible reason for disease susceptibility and resistance in these two clones respectively.

3.1.2. Corynespora leaf disease

A resistant (GT 1) and susceptible (RRII 105) clone were challenge inoculated with *Corynespora cassiicola* and leaves were collected at different time intervals. RNA was isolated from leaves of plants after challenge inoculation as well as from uninfected control plants. RNA sequencing was performed and transcriptome data was generated. Various bioinformatics tools were employed to investigate expression of genes in unchallenged healthy plants as well as pathogen-challenged resistant and susceptible plants. The study provided understanding about the differentially regulated genes and critical pathways involved at different stages of disease development at varying time points. Up-regulation of disease resistant protein and defence-related gene expression ascertained about clonal response to pathogen. Up-regulation of chitinase, beta-glucanase, and genes involved with proteolysis proved ability of the resistant host to degrade pathogen cell wall components. Plant transcription factor (TF) group were up-regulated, which is associated with enhanced immunity against the pathogens. WRKY gene family is among the largest families of TFs in higher plants and are involved in several biological processes such as growth and development, signal transduction, and plant defence against stress. Expression analysis revealed diverse patterns and differential modulation of *Hevea* WRKY gene family in both resistant and susceptible clones in healthy and challenged conditions.

3.1.3. Colletotrichum leaf disease

Effort was taken to isolate, identify and characterize fungal and bacterial endophytes that could be used as antagonistic organisms against *Colletotrichum* spp. for management of Colletotrichum Leaf Disease. Tissue samples were collected from leaves of *Colletotrichum* tolerant and susceptible clone. A total of 44 morphologically diverse bacterial and three fungal endophytes were isolated and purified. All isolates were subjected to dual culture technique in order to identify effective endophytes possessing antagonistic activity against *Colletotrichum* spp. Two bacteria showed up to 47 per cent inhibition and one fungal endophyte grew over the pathogen. In order to confirm the identity of potential endophytes, DNA was isolated from them; PCR amplified for 16S rDNA (for bacteria) and ITS region (for fungi) and sequenced. Bacteria identified were *Burkholderia cenocepacia* and *Ochrobactrum anthropic* and the fungal endophyte was *Trichoderma* spp. All these three organisms have been reported as effective biocontrol agents. Studies dealing with biochemical characterisation and biocontrol activity are in progress.

4. Genome Wide Association Mapping Studies as potential tools to discover regions of disease resistance in rubber genome

Genome Wide Association Studies (GWAS) were initiated by using a collection of 200 Wickham clones. Disease resistance potential was assessed for all these clones to three major pathogens: *Phytophthora meadii*, *Corynespora cassiicola* and *Colletotrichum acutatum* through *in vitro* challenge inoculation experiments. Tolerance to *Phytophthora meadii* was estimated by detached leaf disc assay using zoospore suspension. Tolerance to *Corynespora cassiicola* and *Colletotrichum*

acutatum was evaluated through leaf wilt bioassay using toxin isolated from the respective pathogen. The extreme phenotypes (highly resistant and highly susceptible) were selected for each of the pathogen and six association panels were created (two each for three pathogens) consisting of 11 to 14 individuals in each pool. Equal concentration of genomic DNA from each clone belonging to an association panel was pooled to get a total concentration of 10 µg. Use of a pooling strategy was preferred as it not only reduces the number of samples to be genotyped, but also has the potential to enrich for rare alleles and augment allele effects by extreme phenotypic selection. Genomic libraries were constructed and sequenced using Illumina sequencer (150 x 2 chemistry). The reads were adapters clipped and high quality reads were assembled. Gene prediction was performed for assembled genomes. The adapter clipped reads were further used for alignment against reference genome and variants were called and annotated. The consensus genome was generated from filtered variants.

We have earlier constructed linkage maps, which had 18 linkage groups, reflecting the haploid chromosome number of *Hevea* (n=18) and detected significant QTLs for *Phytophthora, Corynespora* and *Colletotrichum*. Studies on association mapping were initiated to analyse marker-trait associations and QTLs linked with disease resistance. Linkage mapping and association mapping are often applied in conjunction to validate the QTLs identified. Basically QTL analysis has been performed in constructed bi-parental populations using contrasting parents, wherein only one recombination event has been recorded in the population. In our study with association mapping, we have considered use of natural populations or association panels with diverse cultivars with the purpose of recording more recombination events. This will contribute to a higher resolution to find regions associated with traits and serve as a tool to mine the elite genes by structuring natural variation present in a germplasm. The association mapping study will help precisely identify and authenticate putative or reliable markers/ QTLs linked to disease resistance thereby accelerating the pace of disease resistance breeding in rubber trees.

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Project title: Quantitative Trait Loci (QTL) mapping for analysing genetic determinism of disease resistance and development of DNA marker-based selection tools for resistance breeding in rubber tree (*Hevea brasiliensis*).

PLANT PHYSIOLOGY DIVISION

Plant Physiology Division is mainly focusing studies on physiology of rubber plant's growth and yield, environment and stress physiology, screening rubber clones and wild accessions for drought and cold tolerance, gene expression analysis in relation to rubber biosynthesis and ethylene biosynthesis and signaling

1. Environmental and stress physiology

1.1. Studies on adaptive mechanisms in *Hevea* for drought and cold stresses

The experiment was continued to study the adaptive mechanisms of rubber plants for drought and cold stresses. *Hevea* clones with contrasting responses to drought and cold stresses were raised at RRII experimental farm and were subjected to the stress condition. Leaf and stem samples of cold stress exposed and drought treated plants were processed for biochemical parameters and antioxidant enzymes assay to evaluate adaptive responses to withstand the adverse conditions. Under drought and cold condition alteration of pigment composition was varied and decline was in accordance with extension of stress. From the pattern of clonal difference it was noticed that the reduction in pigments level was lesser in SCATC 88/13, RRIM 600 and RRII 208. Variation in leaf water loss (leaf water loss per unit initial water content) was relatively less in clone RRII 208 than other clones. Leaf area was high in RRII 430 and RRII 429 where as RRIM 600 and SCATC 88/13 on par with each other. Biomass partitioning under abiotic stress conditions showed that clone RRII 208 and RRII 429 have a similar trend in biomass allocation. The Root Mass Fraction (RMF) was higher in SCATC 88/13, RRIC 100 whereas high stem mass faction (SMF) was found in RRII 208, RRII 429 followed by SCATC 88/13.

1.2. Screening for cold/ low temperature tolerance potential in germplasm lines through physiological and biochemical approaches

Variability of germplasm lines responses to low temperature/cold stress helps to widen the genetic bases in improving cold tolerance in breeding programs. In this context screening the germplasm accessions and experiment was initiated. Budding of 12 elite clones and 33 genotypes (germplasm accessions) were carried out at CN, Karikkatoor for raising a new polybag nursery in RRII farm for screening for cold tolerance. The budding success for accessions ranged from 15 - 90 per cent whereas 65 - 98 per cent recorded for elite clones. A polybag nursery comprised of 33 germplasm accessions and 12 check clones was raised at RRII farm. Sprouting success was evaluated in check clones and 33 germplasm accessions under stress free period. From the observation under nursery condition it was found that 3 accessions had above 80 per cent and 13 accessions had above 70 per cent success. Among the check clones Tjir 1 showed low sprouting success than other clones. Analysis of early growth indicators like stem development, height and leaf number etc in young plants of 33 germplasm accessions revealed that accessions namely, 102/2003, 287/2003, 218/2003, 521/2004 and 585/2003 were on par with the check clones RRIM 600 and RRII 208. Relative plasticity was worked out in the trial using early growth indicators. It was found that four elite clones (RRIM 600, RRII 429, HN1 and SCATC 98/114) and seven accessions (287/2003, 102/2003, 32/2003, 216/2003, 585/2003,

521/2004 and 19/2003) were on par with stress tolerant elite clones for phenotypic plasticity. Higher degree of plasticity was observed in the clone RRII 208 and accessions 10/2003 and 564/2003. Leaf samples were collected to analyze the allocation of resources and other biochemical parameters are in progress.

1.3. Studies on drought effects on *Hevea* in relation to oxidative stress and antioxidant responses

The study was continued with germplasm accessions (MT 5100 and MT 4788) and *Hevea* clones such as RRII 208, RRII 429 and SCATC 88/13. Polybag plants were raised at RRII experimental farm. Growth performance of germplasm accessions was on par with the check clones. Drought was imposed by withdrawing irrigation for two weeks. The antioxidant enzyme, peroxidase activity was analysed in germplasm accessions such as MT 4788, MT 5100 and the check clones. The data indicated that activity was high in MT 5100, RRII 208 and SCATC 88/13 under drought condition. Analysis of other biochemical parameters particularly xanthophyll pigments, oxidative stress responses and antioxidants are in progress.

1.4. Physiological adaptation of selected ortets under varying agro-climatic conditions in India

A multi-location trial with 16 ortets selected from five different agro-climatic regions of India along with seven check clones were planted at three different locations in 2012 is continued in which CES, Chethackal being one of the locations. The young plants were allowed to grow in a closed planting design till four years, after that alternative plants were removed to allow normal spacing for the plant growth. Mean girth recorded for ortets during March 2021 ranged from 30.1 cm for ortet NGK 69 to 51.7 cm for RRSA 98 (Agartala selection) which was on par with check clone RRII 430 (52.0 cm). Among the check clones RRII 430 and RRII 417 showed better girth increment and attained tappable girth in the eighth year of planting whereas among the 16 ortets, the Agartala selections namely, RRSA 98 (attained tappability) and RRSA 585 recoded better trunk girth than other ortets.

2. Production Physiology (growth and yield)

2.1. Productivity enhancement of NR through high density planting (HDP) and growth regulation by application of Paclobutrazol (PBZ)

A long-term project for the enhancement of productivity of natural rubber through high density planting and growth regulation by application of paclobutrazol (PBZ) is continuing. High density planting (HDP) and application of PBZ indicated that there was a decrease in annual growth of the plants with increasing planting density than normal planting density irrespective of PBZ application. The average girth of trees in D1P1, D1P2, D1P3 and D2P1 (HDP + zero level of PBZ) was significantly greater than that of other treatments with polybag planting material. In case of root trainer plants, the treatments D1P1 and D1P3 have better girth than other treatments. A significant interactive effect of density and PBZ application was found in root trainer plants whereas there was no significant interaction with polybag plants.

2.2. Intercropping with tree crops in rubber

This project was continuing at CES, Chethackal with the major objective of

finding out the impact of tree intercrops and competition for light on the growth and yield of rubber plants. The mean trunk girth was 73.5 cm, 73.2 cm and 72.8 cm for pure stand of rubber, three rows of mahogany and rubber and one row of mahogany trees along with one row of pathimugam trees with rubber, respectively. The growth data indicated that tree intercrops did not affect the girth of rubber plants. The trees were tapped for seven years under S/2 d3 system of tapping and from April 2017 onwards the tapping system was changed to S/2 d7 and monthly stimulation was provided at 2.5 per cent ethephon. The result showed that tree intercrops obstructed rubber yield markedly till 2019-20 but during the reporting year the yield was on par with each other and did not recorded any yield reduction in tree intercropped plots. Among the intercrops, the pathimugam stand was very poor due to shading by the mature rubber trees, whereas mahogany trees were growing better as an inter-crop with rubber because the mahogany canopy height was as equal to rubber canopy in a grown up plantation.

2.3. Effect of stimulation on latex regeneration mechanism in *Hevea brasiliensis*

Experiment on stimulation induced changes in latex regeneration mechanism was continued at CES Chethackal in different clones to study the biochemical and molecular mechanisms associated with latex regeneration after ethephon stimulation. Latex yield, biochemical components and enzyme activities related to oxidative stress (glutathione redox cycle) and ethylene responsive and ROS scavenging related genes were analyzed in soft bark tissues of control and stimulated trees of clones RRII 105, PB 217, PB 260, RRIM 600, Tjir 1, RRII 33 and RRII 38. The trees were under d3 tapping system with three stimulations per year. Increased glutathione content and glutathione reductase and glutathione peroxidase activities were observed in stimulated trees of clone RRII 105 and PB 217. Fifty per cent of the stimulated trees of clone PB 260 became TPD after 4 years of ethephon stimulation. An increased thiol metabolism and low protein and enzyme activities were observed in partial dry trees of clones PB 260.

Expression analysis of ethylene biosynthesis, signaling and ROS scavenging related genes in bark samples of control, stimulated and TPD trees (from stimulated group) of clone PB 260 was also analysed. Ethylene induced TPD trees showed a higher expression of ethylene biosynthetic genes (S-adenosyl methionine synthase, ACC synthase and ACC oxidase). Expression of ROS scavenging related genes (SOD and peroxidase) and some genes related to latex metabolism (glutamine synthatase and ATP ase) were down regulated in these trees.

2.4. Molecular and biochemical basis of ethylene induced latex production in *Hevea brasiliensis* - Ethylene receptors and signal transduction mechanism

To study the molecular mechanism of ethylene induced latex production, comparative expression analysis of five receptor genes (ETR1, ETR2, EIN2, EIN3 and ERF) were carried out in seven clones with different yield potentials (RRII 105,PB 217, PB 260, RRIM 600, Tjir 1, RRII 33 and RRII 38). The mean expression level for each gene in different clones in the bark revealed that ETR1 (ethylene receptor 1) and ERF (ethylene response factor) gene had the highest expression level compared to other

Table Phy. 1 **Relative quantification (fold change) of receptor genes in bark samples of different clones with varying latex metabolism (clone RRII 33 as calibrator)**

Clones	Genes				
	ETR1	ETR 2	EIN 2	EIN 3	ERF
RRII 33	1.0^{cd}	1.0^{c}	1.0^{a}	1.0^{a}	1.0^{c}
RRII 105	1.9^{c}	1.3^{b}	0.67^{bc}	0.99^{a}	2.1^{b}
PB 217	2.7^{a}	0.78^{cd}	0.32^{c}	1.1^{a}	3.2^{a}
PB 260	0.76^{de}	0.97^{c}	0.76^{b}	0.38^{c}	0.83^{c}
RRIM 600	1.1^{c}	2.98^{a}	0.68^{bc}	0.18^{d}	1.0^{c}
Tjir 1	0.55^{e}	0.57^{d}	0.51^{cd}	0.56^{b}	0.89^{c}
RRII 38	0.76^{de}	0.76^{cd}	0.46^{de}	0.26^{d}	0.63^{c}

genes. Between clones highest expression level of these two genes were observed in clone RRII 105 and PB 217 compared to other clones (Table Phy. 1) The receptor genes ETR1 and ERF was also highly up- regulated in these clones after ethylene stimulation. Clone RRII 33 was used as calibrator.

2.5. Studies on inhibitors of ethylene biosynthesis and signalling

To study the regulation of ethylene receptor inhibitors in latex flow and production, trees from clones RRII 105, RRII 430 and RRII 417 were selected and yield recording was continuously carried out for the application of ethylene inhibitors (Silver thiosulphate (silver can be incorporated into receptors instead of Cu co-factor) and 1-Methyl cyclopropene (competitive inhibitor and more affinity than ethylene for the receptor). Ethephon treated trees and unstimulated trees were selected as controls.

2.6. Relationship of ATP status of latex with rubber yield

Field trial with clones/selections planted at RRII (five selections screened based on latex ATP content and three control clones (RRII 105, RRII 417 and RRII 430) was continued. Test tapping yield was recorded during peak yielding season of 2020.

3. Secondary metabolites

3.1. Water relation of latex with reference to the content of inositols and sugars in latex during drought

The trial was continued with selected elite clones to study the relationship of latex osmolytes and water relations. Growth performances of the plants were recorded by taking girth data during peak yielding and stress periods. RRII 430 and RRII 417 observed to have better trunk girthing. Water relation studies of latex during the summer season was carried out in five rubber clones from a trial at Chethackal. The serum osmotic concentration found to have same trend as in the case of peak yielding season in four clones except in clone RRIM 600 which had better osmotic components than other clones studied.

LATEX HARVEST TECHNOLOGY DIVISION

The Division was active in the research and advisory services on all aspects of crop harvesting of rubber. Low frequency weekly tapping is getting acceptance among growers. In the present scenario of shortage of skilled tappers adoption of weekly tapping and controlled upward tapping for old and senile trees will empower the growers. All the programmes progressed well during the period under report. Other activities of the Division included testing and evaluation of various products, advisory and training on all aspects of crop harvesting of rubber.

1. Low frequency tapping

1.1. Programme on popularising weekly tapping

The growers who have participated in the programme continued weekly tapping with satisfactory results. Tapping days realized showed considerable variation due to climatic constraints and practices followed (Tables LHT 1-3).

Table LHT 1. **Yield performance of clone RRII 105 under weekly tapping**

Location/Region	Mean yield ($g\ t^{-1}\ t^{-1}$)	Number of plots
Muvattupuzha		
1	78.8	5
2	96.6	10
3	89.6	2
4	86.1	3
Thodupuzha		
1	75.7	6
Mannarkkad		
1	93.4	3
Thalaserry		
1	143.2	1
Mean	86.7	-
Total	-	30

Table LHT 2. **Yield performance of clone RRII 414 under weekly tapping**

Location/Region	Mean yield ($g\ t^{-1}\ t^{-1}$)	Number of plots
Kottayam		
1	91.6	2
Muvattupuzha		
1	77.9	1
2		3
Thodupuzha		
1	115.6	3
Thalaserry		
1	68.5	1
2	105.6	2
Mannarkkad		
1	72.6	2
Mean	88.6	-
Total	-	16

Table LHT 3. **Yield performance of clone RRII 430 and mixed clone under weekly tapping**

Location/Region	Mean yield ($g\ t^{-1}\ t^{-1}$)	Number of plots
Kottayam		
1	79.8	3
Muvattupuzha		
1	124.8	2
2	89.5	2
3	105.6	4
4*	111.5	1
Kottarakkara		
1	106.9	1
Palakkad		
1	86.3	1
Mannarkkad		
1	85.8	1
Belthangady (Karnataka)	92.8	05
Mean	98.1	-
Total	-	20

*Mixed clone

1.2. Low frequency (d10) tapping system in clone RRII 105

1.2.1. Large scale trial on d10 frequency of tapping in clone RRII 105

The large scale onfarm commercial evaluation trial in 1987 field on d10 frequency tapping initiated during 2015-16 at Kanthimathy Estate , Kulasekharam, Tamil Nadu in 10 tapping blocks was continued. Mean dry rubber yield of 1794 kg ha^{-1} with 30 tapping days on renewed basal panel tapping could be obtained under d10 frequency of tapping during 2020-21. The g $t^{-1}t^{-1}$ ranged from 50 to 223 g . With the introduction of Controlled Upward Tapping further yield increase could be obtained. The g $t^{-1}t^{-1}$ under CUT ranged from 62 to 245 g (Table LHT 4).

Table LHT 4. **Monthly variation in yield performance of clone RRII 105 in d10 frequency of tapping at Kanthimathy estate, Kulasekharam**

Month	kg block $^{-1}$	g t^{-1} t^{-1}	kg block $^{-1}$*	g t^{-1} t^{-1}*
Apr'20	50	50	64	62.3
May	82	82	82	83.1
June	110	138	119	129.4
July	157	147	194	178.1
Aug	152	166	150	165.8
Sep	218	203	231	212.4
Oct	222	209	270	244.6
Nov	238	223	244	220.7
Dec	210	187	222	220.0
Jan'21	155	156	214	194.6
Feb	129	130	134	145.9
Mar	71	66	71	63.5
Total	1794		1995	
Mean	149	146	166	160

*With CUT

1.2.2. Exploratory trial on d10 frequency of tapping in clone RRII 105

This exploratory trial on d10 frequency of tapping was initiated at CES, Chethackel in field 1987 in clone RRII 105. Controlled Upward Tapping (S/3 d10) was practiced during non rainy months and rest of the months trees were tapped in the basal panel tapping (S/2 d10). Yield of 36 kg t^{-1} and 91 g $t^{-1}t^{-1}$ were obtained during 2020-21.

1.2.3. Large scale experiment on Low Frequency Tapping (d10) in clone RRII 105 (Panel BO - 1)

The large scale experiment on d10 frequency of tapping in comparison with weekly tapping to study the yield performance of clone RRII 105 in 2009 field, laid out during 2018 -19 at Kanthimathy Estate, Kulasekharam, Tamil Nadu was continued. There were five treatments comprising of weekly and d10 frequencies of tapping and different levels of stimulation. Significant yield variation among the treatments was observed. Higher dry rubber yield was observed under weekly tapping. Per tree yield of 5 kg could be realized under d10 frequency of tapping with ethephon (ET 2.5%, 5 % Pa 18/y) application (Table LHT 5).

Table LHT 5. **Yield performance of clone RRII 105 under d10 frequency of tapping**

Treatment	g t^{-1} t^{-1}	kg tree $^{-1}$
T1- S/2 d6 ET 2.5% Pa 12/y	105.3 c	5.6 a
T2- S/2 d6 ET 5% Pa 12/y	109.7 c	5.8 a
T3- S/2 d10 ET 2.5% Pa 18/y	140.1 ab	4.8 bc
T4- S/2 d10 ET 5% Pa 18/y	130.1 b	4.4 c
T5- S/2 d10 ET 2.5%, 5 % Pa 18/y	149.2 a	5.0 b

Values followed by same letter/s are significantly different

1.2.4. Large scale experiment on Low Frequency Tapping (d10) in clone RRII 105 (panel BO-2)

An RBD experiment was laid out in six blocks of clone RRII 105 (Field 2002) and imposed yield stimulation as per the schedule under LFT systems. There were six treatments comprising d7 (with monthly stimulation as control) and d10 frequencies of tapping with different frequencies of

stimulation. Yield under weekly tapping with monthly stimulation (ET.2.5%) was comparable to that of d10 frequency of tapping with 5 per cent ethephon (18/y). stimulation (Table LHT 6).

Table LHT 6. **Yield response of Low Frequency Tapping (S/2 d10) in clone RRII 105**

Treatment	Yield (kg 400 trees^{-1})
T1- S/2 d7 ET 2.5% 12/y	1240 a
T2- S/2 d10 ET 2.5% 36/y	646 b
T3- S/2 d10 ET 2.5% 18/y	702 b
T4- S/2 d10 ET 5% 18/y	923 ab
T5- S/2 d10 ET 2.5% 12/y	807 b
T6 – S/2 d10 ET 5% 12/y	614 b

Values followed by a common alphabet are not significantly different

2. Controlled Upward Tapping (CUT)

2.1. Large scale on farm trial on Low Frequency Controlled Upward tapping (LFCUT) under weekly tapping

The large scale on farm trial on Low frequency controlled Upward tapping (LFCUT) under weekly tapping with periodic panel change initiated during 2017 at Kanthimathy estate, Kulasekharam, Tamil Nadu in 12 tapping blocks (8 blocks of 1978 mixed clone and 4 blocks of 1962 seedling population) was continued with promising results and the system seems promising for further reduction in cost of production of NR for mixed and seedling population also.. (Table LHT 7)

Table LHT 7. **Yield performance of Low Frequency Controlled Upward Tapping (LFCUT) under weekly tapping**

	Mixed clone		Seedling population	
Month	kg block^{-1}	g t^{-1} t^{-1}	kg block^{-1}	g t^{-1} t^{-1}
Apr'20	62	36	79	52
May	58	40	82	52
June	97	66	129	81
July	135	81	154	95
Aug	152	89	91	95
Sep	151	92	181	89
Oct	186	98	122	93
Nov	169	95	201	124
Dec	190	97	233	123
Jan'21	185	111	182	120
Feb	148	79	137	92
Mar	107	60	148	72
Total	1634	-	1737	-
Mean	157	79	145	91

3. Other experiments

3.1. Response of RRII 400 series clones to yield stimulation

At CES, Chethackal, in field 2004, RRII 400 series clones (RRII 414, RRII 422 and RRII 429) were identified for the experiment. The statistical design was completely randomized single tree single plot. Tapping system adopted in this trial was S/2 d3 6d/7. Three rounds of stimulation (ET 2.5% pa) was given during 2020-21 in comparison with the unstimulated control trees (50% trees for stimulation and 50% trees unstimulated, for each clone).

In clone RRII 422, stimulated trees showed significantly higher yield than unstimulated trees. No significant yield increase was noticed in stimulated trees of clones RRII 414 and RRII 429 (Fig. LHT 1). Good yield was observed in both stimulated and unstimulated trees of clone RRII 422 than other two clones of RRII 414 and RRII 429 in BO-2 (5) panel.

3.2. Response of clone RRII 430 to yield stimulation under LFT

In another experiment at HML, Palapilly Estate, performance of clone RRII

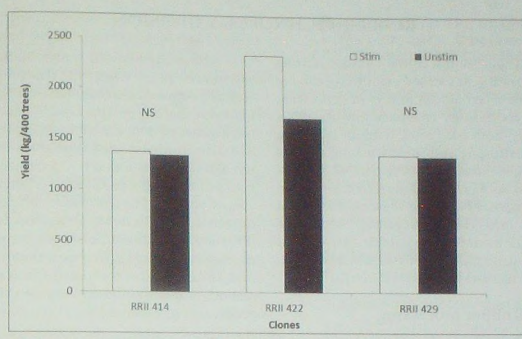

Fig. LHT 1. Yield performance of RRII 400 series clones

430 to yield stimulation under Low Frequencies of Tapping (d3, d4 and d7) was studied in field 2000 (Panel BO-1). There were four treatments comprising of d3 (without stimulation as control), d3 (2/y), d4 (4/y) and d7 (12/y) frequencies of tapping.

Table LHT 8. **Yield response of clone RRII 430 to low frequency tapping**

Treatment	Yield ($g\ t^{-1}t^{-1}$)
T1- S/2 d3	72.8 c
T2- S/2 d3 ET 2.5% 2/y	77.7 bc
T3- S/2 d4 ET 2.5% 4/y	86.2 b
T4- S/2 d7 ET 2.5% 12/y	112.0 a

Significant yield variation among the treatments were observed. Mean dry rubber yield of 112 g $t^{-1}t^{-1}$ was observed under weekly tapping as against 73 g $t^{-1}t^{-1}$ under d3 frequency of tapping with out stimulation. Dry rubber yield significantly as high as 112 g $t^{-1}t^{-1}$ could be observed during 2020-21 in spite of agro climatic and other constraints which indicate feasibility of weekly tapping in clone RRII 430 (Table LHT 8).

4. Testing and evaluation of products, training and advisory

Many rainguard adhesive samples, LDPE samples and Ethephon samples were tested during the year under report. One model of mechanised (motorized) tapping machine was tested and evaluated by the Division and approved as a tool for basal panel tapping of rubber trees. Testing fee as part of IEBR, for testing of various products could be collected.

Under Sasthradarsan Programme, growers were given exposure to various aspects of crop harvesting of rubber. Through the in-house training programme, detailed theoretical and practical aspects of latex harvest technology were also provided to trainees/growers. Besides, advisory services on crop harvesting and allied aspects were also extended to the growers during the year under report

RUBBER TECHNOLOGY DIVISION

The activities of the Division during the current year focused mainly on evolving improved techniques in rubber processing latex technology (preparation of DPNR directly from filed latex without creaming or centrifuging and new leaching process to reduce extractable protein in NR surgical gloves), rubber technology (NR latex carbon black master-batch and silica reinforcement of NR) and rubber recycling (stable free radical assisted devulcanisation and cytotoxicity analysis of crumb rubber from end of life tyres).

1. Rubber processing

1.1. A study on the effect of storage on raw rubber properties of cuplumps

A study was conducted on the effect of storage on properties of cuplumps especially on plasticity retention index (PRI). Raw rubber properties of cuplump as function of storage time is shown in Fig. Chem. 1.

The study showed that initially the raw rubber properties of the cuplumps as well as the mixed field coagulum were good. However there is an appreciable quantum of microbes present especially in cuplumps and also in mixed coagulum even in the initial time. It was found that the microbial populations are not subsided even after one year of storage. The raw rubber properties especially the Plasticity Retention Index (PRI) are also falling below the required standards on storage. Therefore the storage and transportation of cuplumps with respect to quality and in view of human health are not encouraging.

1.2. Process development for skim latex processing

The development of a new and efficient process for complete recovery of high quality skim rubber was reported with the skim coagulant SC-C. The skim latex coagulants (SC-1 and SC-2) were procured from two local sources and its comparison

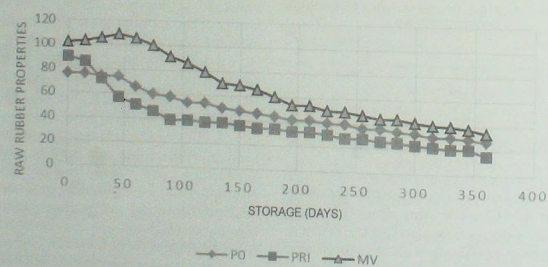

Fig. Chem. 1. Raw rubber properties of cup lump as a function of storage time

with the control skim coagulant (SC- C) was carried out in the reporting period. The skim coagulant SC-1 was found to be effective in complete recovery of skim rubber like the control skim coagulant, whereas SC-2 was not effective. The activity of SC-1 was found to be 80 per cent to that of the control skim coagulant (SC-C).

1.3. Development of low cytotoxic NR latex products

NR latex products possess high residual chemical contents and this leads to high cytotoxicity and low bio-compatibility. In house balloon formulations F3 and F4 with NR cenex and other NR latex compounding ingredients were prepared by varying the sulphur to accelerator ratio. The film were prepared from both pre-vulcanised latex and direct latex compounding by dipping and then cured at 100 °C for 1 hr. The residual accelerator content was quantified as copper-accelerator complex by UV spectroscopy at varying experimental conditions (Table Chem. 1).

1.4. Profiling of fungal toxins from *Corynespora cassiicola*

The work was done in collaboration with the Pathology Division of RRII. The purification of the secondary metabolite toxins from the serum of *Corynespora cassiicola* isolated at RRII was done using the flash chromatography. Four active toxin fractions were identified by leaf toxin assays. Further purification of the active toxin fractions are under progress.

1.5. Establishing testing of imported rubber products laboratory and REACH compliance laboratory

A project report for establishing testing facility for imported rubber products was prepared in consultation with the Technical Consultancy Division and submitted. The procurement of GC-MS MS Triple quadrupole and ICP-MS for the REACH compliance laboratory was done in consultation with the Technical Consultancy Division through GeM bidding.

2. Rubber technology : Reinforcement of rubber

2.1. NR latex carbon black master-batch

Studies on the preparation of NR latex - carbon black master batch were continued. Latex-carbon black masterbatches were prepared in presence of non-ionic, anionic and cationic surfactants. Also attempted a reverse mechanism for preparation of latex-carbon black masterbatches.

2.1.1. Technological properties of NR latex-carbon black masterbatches prepared in presence of a non-ionic surfactant

The samples were compounded as per the formulation given in Table Chem. 2 and the vulcanizate properties were studied and compared with a control (Dry mix). The results are shown in Table Chem. 3.

The test results of carbon-black masterbatches with the non-ionic surfactant shows that most of the technological properties improved with the use of the new surfactant.

Table Chem. 1. **Residual accelerator content**

Formulation	Accelerator to sulphur ratio	Residual accelerator in microgram/g		
		With leaching	Without leaching	Pre vulcanised latex
F3	0.75 : 1.25	62	60	30
F4	0.75 : 1.50	70	85	32

Table Chem. 2. **Formulation of the mixes**

Ingredients	Phr (g)
Master batch	(Effective NR 100g)
ZnO	4.5
Silica	8
Stearic acid	3
TMQ	1
6 PPD	2.75
TBBS	1.6
Sulphur	1.2

Table Chem. 3. **Vulcanizate properties of masterbatch with non-ionic surfactant**

Tests	Control	M.B.
Tensile strength in (Mpa)	24.0	23.0
Tear strength in N/mm	135.0	128.3
Elongation at break in (%)	574.0	603.0
Modulus @100%	2.9	2.8
Modulus @200%	6.2	5.8
Modulus @300%	10.6	10.3
Hardness (Shore A)	62.0	69.0
Compression Set in (%)	33.9	35.7
Heat buildup	22.0	27.0
Abrasion resistance	117.6	125.0
Flex resistance (Cycles)	58493.0	116319.0

Table Chem. 4. **Technological properties**

Tests	Control	MB (SDS)
Tensile strength (Mpa)	18.7	21.9
Elongation at break (%)	693.3	704.4
Modulus @100%	1.2	1.3
Modulus @200%	2.2	2.4
Modulus @300%	3.8	4.2
Modulus @500%	9.0	10.3
Tear strength in N/mm	40.4	44.0
Hardness (Shore A)	4	50.0
Compression Set (%)	38.0	35.5
Heat buildup	12.0	11.0
Abrasion resistance	195.8	159.8
Flex resistance (Cycles)	616638.0	639274.0

2.1.2. *Technological properties of NR latex-carbon black masterbatches prepared in presence of an anionic surfactant*

The results showed that with the anionic surfactant tensile strength, modulus, EB, tear strength and hardness increased compared to control. Heat buildup and abrasion resistance also improved with the new surfactant. Though flex resistance is more with the masterbatch, the improvement is not considerable. It was also found that for this particular latex the values are lower even for the control (dry mix). However, all the properties of the masterbatch were better compared to control (Table Chem. 4).

2.1.3. *Technological properties of NR latex-carbon black masterbatches prepared in presence of a new cationic surfactant*

Table Chem.5. **Technological properties**

Properties	Control	MB
Tensile strength (MPa)	27.1	21.4
Tear strength (N/mm)	128.8	140.4
Heat build up (°C)	22.0	27.0
Abrasion loss (mm^3)	72.6	70.0
Modulus @100 %	2.8	4.4
Modulus @200 %	6.9	10.3
Modulus @300 %	12.5	16.7
EB	565.8	388.9
Compression set (%)	22.7	22.4
Flex resistance (Cyles)	66638.0	163687.0

The results showed that tear strength, and modulus and flex resistance were increased. Heat buildup was inferior (Table Chem. 5).

2.1.4. *Reverse mechanism*

Latex carbonblack masterbatches were made using a reverse mechanism. The technological properties are shown in Table Chem. 6.

The results showed that the masterbatch prepared by the reverse mechanism shows better properties except compression set. Other properties are better or on par with the control. However, all the values are lower than expected. The control

Table Chem. 6. **Technological properties**

Tests	Control	MB
Tensile strength in(MPa)	18.17	22.03
Elongation at break in (%)	358	356
Modulus @100%	3.92	5.36
Modulus @200%	8.74	11.59
Modulus @300%	13.34	18.60
Tear strength in N/mm	105	147
Hardness(Shore A)	64	70
Compression Set in (%)	33.5	43.44
Abrasion resistance	107	108
Flex resistance (Cycles)	104712	216705

also showed low values. This may be because the latex collected during this season may not be good.

2.2. Silica reinforcement of NR

In continuation of the work carried out on effect of cure system on the coupling efficiency of silanised silica (sSilica) a number of various cure systems were analysed. Table Chem. 7 gives the result of some of the promising cure systems which showed excellent vulcanisate properties including abrasion loss in comparison with the HAF filled NR.

2.3. NR polymeric filler

In the present study blends of natural rubber were blended with low proportions of high density polypropylene. The objective was to improve the properties of natural rubber vulcanisate by blending with low proportions of high density polypropylene.

Blending of natural rubber and HDPE was carried out in Haake Rheocord 90 at 160 °C for 10 minutes at 60 rpm. The formulation of the compounds is given in the Table Chem. 8. The rubber compounds were prepared in a laboratory open two roll mill having friction ratio 1:1.25. The rubber compounds were prepared in ambient temperature. Blend ratio of NR:HDPE selected was 100:0, 90:10, 80:20 and 70:30.

Table Chem. 8. **Formulation mixes**

Ingredients	Control	NR: HDPE 90:10	NR: HDPE 80:20	NR: HDPE 70:30
NR	100	90	80	70
HDPE	0	10	20	30
Zinc oxide	5	5	5	5
Stearic acid	2	2	2	2
HSL	1	1	1	1
CBS	1.5	1.5	1.2	1.5
Sulphur	1.5	1.5	1.5	1.5

The cure characteristics of the compounds were studied using RPA 2000. Cure time, scorch time, minimum torque and maximum torque values were recorded for this test. In this study, minimum torque decreased with increase in the HDPE content in the blends. This may be due to the melting of un-crosslinked HDPE in the blend during curing. Cure test was carried out at 150 °C which was higher than the melting temperature of HDPE.

Table Chem. 7. **Comparison of cure system in NR/sSilica reinforcement**

Property	CBS/DPG/S	Cure system4	Cure system 5	Cure system 6	NR/HAFSemi-EV
Tensile strength, MPa	25	25.7	29.9	27.4	25
M100, MPa	2.5	3.8	4.2	2.3	2.5
M200, MPa	5.7				6.2
M300,MPa	9.9	13.8	14.8	10.7	10.9
EB,%	565	488	502	542	508
Tear, N/mm	115	115	103	105	107
Din abrasion loss, mm^3	157	97.4	111.4	103	96.3
Hardness, Shore A	70	76	67	70	58

2.3.1. Technological properties

The technological properties of the vulcanisates such as tensile strength, modulus, hardness, tear strength, elongation at break, compression set etc were studied (Table Chem. 9). The tensile strength of the blend with HDPE increased with increase in the HDPE content from 0-30. The modulus at 100 per cent and 300 per cent also showed the same trend as in the case of tensile strength. It can be seen that modulus at 300 per cent improved substantially. Modulus at 300 per cent is a measure of cross link density and it also shows reinforcing effect of the HDPE in the NR vulcanisates. The improvement in the tensile strength and modulus indicates the reinforcing capacity of HDPE in the NR/HDPE blend.

Table. Chem. 9. **Technological properties of the blends**

Properties	Control	NR: HDPE 90:10	NR: HDPE 80:20	NR: HDPE 70:30
Tensile strength, MPa	23.7	25.3	26.2	27.3
M, 300, MPa	1.8	2.8	5.8	6.7
Elongation at break, %	834	769	670	602
Tear strength,N/mm	33	39	42	50
Compression set, %	22	24	30	34
Hardness, Shore A	40	48	58	70
DIN Abrasion loss, mm3	187	130	102	78

Compression set of the NR/HDPE blend was higher than the control compound. This property increased drastically with higher HDPE ratio in the blend. This test was carried out at 70°C for 22 hours. The increase in the set property with more HDPE ratio also support this nature of plastic. DIN abrasion loss indicates the abrasion resistance of the vulcanisates. Lower the abrasion loss higher is its resistance. Blend with higher proportion of HDPE showed low abrasion loss *ie.*, high abrasion resistance. Abrasion resistance increased with HDPE concentration.

3. Development/advisory work/ project work

Tested and report was given for the damaged tyres referred from various consumer disputes redressal forum in the country.

TECHNICAL CONSULTANCY DIVISION

Technical Constancy Division was constituted with the aim to appease the demands of the rubber based manufacturing industries in the country. The goals of the Division are intended in such a way that the rubber based units of the country numbering to over 3845 will be able to take advantage of the applied research and developmental activities being conducted in RRII. Technical Consultancy Division is an NABL approved laboratory and has the facility to test enormous parameters of rubber products. The latex products testing section of the Division has almost all facilities and probably it is the only one laboratory in the public sector. Since the laboratory is following ISO 17025: 2005 norms for its routine analysis its test certificates are universally valid and are valuable to the exporters of rubber products in the country. The Division also stack up knowledge on industrially important problems by conducting R&D programmes so that the problems in the manufacturing

sector can be overcome within the shortest possible time.

The services provided are R&D activities of rubber industry (both products and processes), development of new products, testing/certification of rubber products as per relevant national and international standards. The services offered by the division include (i) testing support to industries as per national and international standards *i.e.*, ISO, BIS, ASTM, EN, ASRTU *etc.* ii) Product development-demonstration/practical training for quality improvement (iii) evaluation of chemicals (iv) preparation of project profiles and technical bulletins (v) advisory services and (vi) conducting awareness meetings/lectures to entrepreneurs regarding trouble shooting/ cost reduction of factory processes.

The highlights of the projects are given below.

1. Research Projects

1.1. Rubber based adhesives: Carbon black filled natural rubber-based solution adhesives: Effect of tackifiers

In this study the performance of a natural rubber (NR)-based solution adhesive for rubber to rubber bonding was evaluated. This work mainly focused on the effect of wood rosin (WD), coumarone-indene (CI), terpene-phenol (TP) and phenol-formaldehyde (PF) resin tackifiers on the vulcanization characteristics, mechanical properties and adhesion strength of carbon black (CB) filled NR-based solution adhesives. Retardation of vulcanization properties was observed by the addition of tackifiers. The addition of tackifiers improved the peel strength of NR-based solution adhesives for rubber-to-rubber bonding (Fig. TC. 1).

1.2. Polychloroprene-based solution adhesives: role of tackifiers on adhesive properties

Solvent-based polychloroprene (CR) adhesives were formulated using four different types of tackifiers. Wood rosin (WD), coumarone-indene resin (CI), terpene-phenolic resin (TP) and para-tert-butyl phenol-formaldehyde (TBPF) resins were incorporated with CR at various amounts say 20 to 50 phr (parts per hundred rubber). The effect of nature and amount of resins on adhesion strength was measured on both leather to leather and rubber to rubber joints. Results (Fig. TC. 2) indicated that the nature and amount of resin greatly affected the performance of the CR adhesive.

1.3. Polychloroprene-based solution adhesives: effect of nanoclays on adhesive properties

The effect of nanoclays on the properties of polychloroprene (CR)-based adhesive was examined. Unmodified montmorillonite clay, Cloisite Na+ (CLNa) and modified nanoclays Cloisite 10A (CL10A), Cloisite 15A (CL15A) and Cloisite 93A (CL93A) were selected for the study. Addition of nanoclays (CLNa$^+$, CL10 A, CL15 A and CL93 A) improved the adhesion strength of CR-based contact adhesives (Fig. TC. 3).

1.4. Polymer blends: carbon black filled natural rubber/butadiene rubber composite

In the present work, various combinations of natural rubber (NR), butadiene rubber (BR) and reclaim rubber (RR) were prepared with a view to produce low cost re-treading materials. The vulcanization characteristics, mechanical properties and thermal properties of the blends were evaluated. The NR rich system

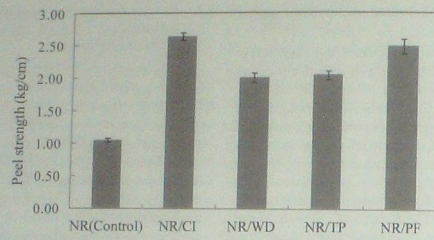

Fig. TC. 1. Peel strength of NR-based solution adhesives (rubber-rubber bonding)

registered good tensile strength and tear values.

In the present work, latex reclaim (LR) was mixed with NR/BR at various proportions to produce tyre retread materials. Results indicated that the scorch time and cure time were decreased with the increase of reclaim loading. The mechanical properties like tensile strength, tear strength and abrasion resistance decreased with the increase in the LR content. It was found that 70-80 per cent of the mechanical properties were retained even after addition of 30 phr of LR.

1.5. Radiation vulcanization of natural rubber latex: Effect of fumed nano silica and nano clay addition on radiation vulcanization of natural rubber latex

The nano and micro composites of radiation vulcanized natural rubber latex

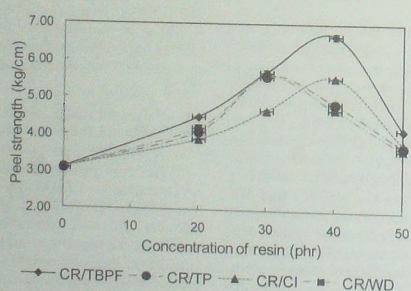

Fig. TC. 2. Peel strength on leather-leather joints

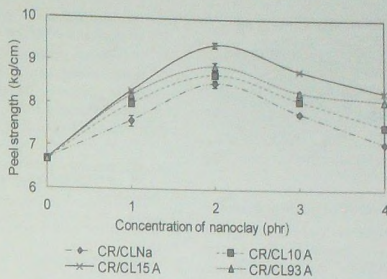

Fig. TC. 3. Peel strength on leather-leather joints

(RVNRL) were prepared by incorporating aqueous dispersion of fumed silica, layered silicate and conventional filler (china clay). The results showed that the nano silica particles were homogenously distributed throughout NR matrix with particles in the size range from 10 to 15 nm. The crosslink density and mechanical properties were analysed and found that RVNRL/ nanosilica composite showed better physical properties compared to others (Fig. TC. 4).

1.6. Transparency of different prevulcanized natural rubber latex and their composites

The effect of different types vulcanization (sulphur, peroxide and radiation) in NR vulcanizaties and their composites on light transmittance was studied (Table TC. 1). Among three different types of prevulcanized latex films, radiation vulcanized natural rubber latex registered the maximum transparency. The addition of

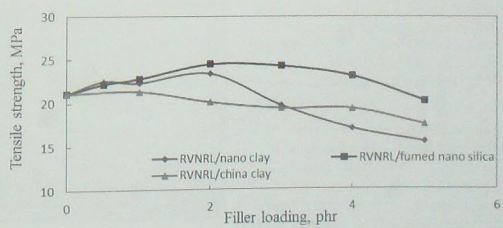

Fig. TC. 4. Tensile strength of RVNRL/fumed nano silica, nano clay and china clay composites

Table TC. 1. **Effect of different prevulcanized latex on transparency**

Sample	Transmittance (%)
RVNRL	71.6
PVNRL	48.3
SVNRL	64.6

nano meter sized fillers like nano silica, layered silica etc is effective to obtain high transparency.

1.7. Shelf life study of medical gloves

The life expectancy of surgical gloves by real time experimentation and accelerated conditions . The experiment was done as per the guidelines of ASTM/ ISO 13320:2005. Effort was taken to establish the effectiveness of the existing Arrhenius and WLF models to evaluate the shelf-life of surgical gloves.

As per the specifications surgical gloves has to maintain 75 per cent retention of properties during the service life. It is observed (Fig. TC. 5) that 75 per cent retention for tensile strength was obtained at 68°C wheras tear strength registered the same at 64°C and force at break at 66°C. Crosslink density values increased as the temperature increases from room temperature to 80°C.

1.8. Technological properties of prevulcanized creamed latex

The major processing parameters of the latex compounds studied were Brookfield viscosity; Total solid content, Mechanical stability time, pH etc. Results are given in Table TC. 2.

Table TC. 2. **Properties of prevulcanized creamed latex (PPFCL) and creamed latex (CL) (control)**

Sl. no.	Parameters	PPFCL	CL (control)
1.	Total solid content (TSC), %	55.5	56.0
2.	PH	10.2	10.4
3.	Brookfield viscosity, cP	207.5	175.5
4.	Mechanical stability time (MST), sec	920	836
5.	Particle size, µm	0.521	0.545
6.	Chloroform number	3	2

In this study, field latex was prevulcanized and subjected to creaming. Results showed that prevulcanized latex produced by creaming of field latex gave films of better quality and such materials gave products with excellent transparency.

2. Rubber Products Incubation Centre (RPIC)

The Rubber Products Incubation

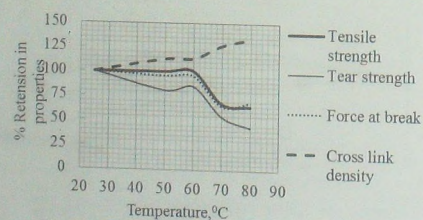

Fig. TC. 5. Effect of temperature on property retention

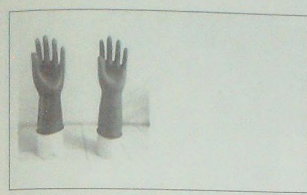

Fig. TC. 6. General purpose gloves prepared form blends of NR: XSBR 80: 20

Fig. TC. 7. Rubber bands prepared from blends of NR: XSBR 80: 20

Centre (RPIC) was commissioned at RRII in June 2020.

The focus of the centre, to incubate and develop innovations and ideas of the entrepreneurs in the rubber industry value chain in to reality through scientific and technological upgradation, upholding the principles of sustainability and protection of environment. Innovative ideas of 8 companies were registered and all the projects are running very well.

2.1. Development of hand gloves and rubber root trainer cups by M/s. EOC Polymers India Pvt. Ltd., Hariyana, India, Development of NR / SBR latex for dipping industry

Fig. TC. 8. Orthotic insoles produced from 100 % natural rubber or

This start-up programme was initiated to produce low-cost high performance gloves from blends of XSBR and NR. The project was further extended to the production of root trainer cups, rubber bands *etc.* Accordingly different blend proportions were prepared with XSBR grades and NR latex. Optimized the cure systems and evaluated the properties. The photographs of the products are given in Figs. TC. 6 & 7.

2.2. Natural rubber latex based products for footwear and lining of artificial limbs by M/s. Profoma (Unit of GLARF Palakkad)

Orthotic insoles for diabetic and leprosy persons were developed using 100% natural rubber. The orthotic insoles developed are very soft (Shore A-15) and has minimum compression set (below 10%). The product has the required density and is cost effective. The clinical trials of the product were successfully completed at Vellore Medical College.

2.3. Low cost latex prevulcanization system by M/s. Royal latex products, Kottayam, Kerala

In this study a suitable cold vulcanization system is proposed for the manufacture of house-hold gloves. Different combinations of accelerated sulphur cure system were tried

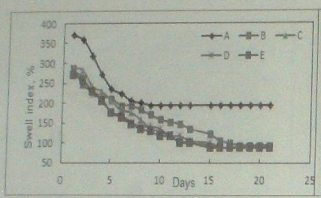

Fig. TC. 9. Change in crosslink density during room temperatire vulcanization

Fig. TC. 10. Handgloves produced from room temperature vulcanization.

under specified temperature. The cure state of the latex was followed through crosslink density measurements. Several trials were conducted and optimized the dose of chemicals and conditions for room temperature vulcanization (Figs. TC. 9 & 10).

2.4. Development of high voltage electrical mats by M/s. Dolphin Rubber Industries, Industrial Development plot, Kottayam

The work was initiated with EPDM rubber incorporated with non-black fillers. In order to impart flame retardancy appropriate fillers were included. The mat compound made accordingly was send for testing at National Test House Chennai for high voltage resistance test.

2.5. Utilization of the rubber tiles from scrap nitrile rubber by M/s. John Traders LLP, Industrial Estate, Changanacherry

The proposal was to make rubber tiles using the scrap of the nitrile rubber gloves. In order to make the process environmentally friendly, a water-based adhesive was used to bind the gloves scrap (Fig. TC. 11). The tiles developed were successfully passed the industrial trials and the know-how was transferred to the client.

2.6. Adhesives for rubber lining of chemical storage tanks by M/s. Adhic Rubber Indiurties, Muttambalam, Kottayam

Very often the rubber compound used for lining work in storage tanks are different

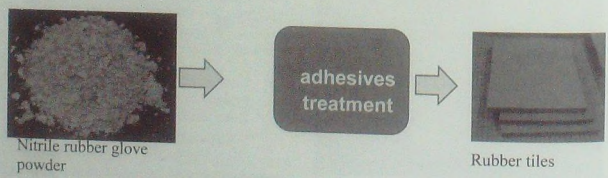

Fig. TC. 11. Process flow chart of rubber tiles manufactured from scrap nitrile rubbers

Fig. TC. 12. Process flow chart for the manufacture of Block rubber from cuplumps

in order to match with the liquid inside the tank. Here the adhesive plays an important role in bonding the rubber based lining material to the metal. Different formulations were tried in order to get better bonding between the rubber based lining material and the metal.

2.7. Production of block rubber with superior raw rubber properties by M/s. Kavanar Latex, Pala

A process modification and use of good quality raw materials are proposed for the production of block rubber. Experiments were conducted in this line and have got block rubber with excellent raw rubber properties (Fig. TC. 12).

2.8. Development of epoxidised natural rubber by M/s. Harrisons Malayalam Plantations, Kochi

Epoxidised natural rubber (ENR) is a chemically modified rubber having superior chemical and air retention properties. Besides ENR can accommodate silica more effectively than NR which has applications in tire and similar products. Prepared ENR 25 and 50 and the characterization of the same is in progress.

3. Rubber product testing and product development

3.1. NABL Accreditation

In order to switch over the NABL accreditation of TC laboratory from the old system (ISO/ISE TC 17025-2015), to the new one (ISO/ISE TC 17025-2017), the quality manual and all other relevant documents were prepared in line with the guidelines of ISO/ISE17025:2017 and uploaded the same to NABL portal. Successfully completed the NABL online audit conducted on 10/10/2020. Accordingly, the Technical Consultancy Laboratory has received the accreditation as per the latest NABL guidelines (ISO/ISE 17025-2017).

3.2. Rubber product testing

Rubber products derived from NR/SR and also from thermoplastics were tested as per the relevant national and international standards (export market). Different types of rubber products tested include bridge bearings, rubber diaphragms, pre-cured/ conventional treads, bonding gum, floor mats, Hawaii soles, sponge rubber, O-rings, bushes, engine mounts, automobile components etc. The major latex products tested include examination and surgical

gloves, latex adhesives, latex thread, balloons, Folly catheters, condoms etc.

The details of the rubber products tested during 2020-21 is given in Table TC 4.

Table TC. 4. **Number of samples tested and the revenue collected (2020-21)**

No. of samples tested	718
No. of clients	403
No. of parameters analysed	2402
Consultancy letters/e-mails	2022
Extractable protein analysis	277
Hands-on training imparted	6
No. of test reports issued	718
Component analysis	58
Total revenue collected, (Rs.)	29,41,451/-

3.3. Rubber product development

The division offers services to entrepreneurs as well as existing rubber based industries for the development of rubber products based on both synthetic and natural rubber. The quantum of product development during the reporting period is given in Table TC. 5.

Table. TC. 5. **Number of know-how transferred to industries**

Name of products developed	Numbers
Adhesives	4
Automobile components	3
Expanded rubber sheets and soles	2
Fire resistant mats	6
Latex based dipped and foamed goods	2
Pre-cured tread, bonding gum and tube valve	3
Rubber based agro machinery components	4
Rubber based engineering components for railway, defence, BSF etc.	2
Rubber moulds	1
Rubber tiles	2
Total	29

3.4. Advisory services

Matters relating to various aspects like selection of raw materials, dosage of ingredients, redesign of formulation, processing conditions, recent regulation etc. were always a subject of concern among the clients. Division has given appropriate guidance in all these aspects.

Some of the major public sector clients who availed the services of the division are VSSC, BPCL, Kochi Metro etc.

4. External funded projects

4.1. Preparation of project reports for setting up of large-scale factories

- Fully automatic surgical gloves manufacturing unit for M/s Ravenbuck Latex & Surgicals, Rubber Park, Irapuram, Kottayam.
- Commissioning of the intermix facility for rubber compounding at Common Facility Service Centre (CFSC), Manjeri
- Manufacture of medical gloves for M/s. Padak Rubbers (P) Ltd, Adoor P.O, Kottayam.
- Manufacture of medical gloves for M/s. Kannur Natural Rubber Products (P) Ltd, Kannur.
- As per the request of the entrepreneurs, project profiles and technical bulletins were issued on payment basis.

4.2. Funded Industrial projects

- Re-designed the automatic dipping plant and also provided the technology for the manufacture of household gloves for NEBEL India Pvt, Bodhjung Nagar, Tripura
- Plant lay-out was offered to M/s. Maximus Rubber Industries, a tread rubber manufacturing unit at Tripura.

- Construction of the dipping plant for the manufacture of disposable latex shoes (patented product) to M/s. DNNG and Company, Mumbai.
- Consultancy to M/s Kerala Minerals & Metals Ltd, Chavara, Kollam for the study on premature failure of rubber lining materials in chemical storage tanks.
- The manufacturing technology for Hawaii soles has been transferred to an entrepreneur, Bangalore.
- Recommendations given to M/s BKT Tyres for overcoming the defects in the rubber compound.

5. Infrastructure development and implementation of new proposals

5.1. REACH compliance laboratory/ Laboratory for imported rubber products

Constructed a new laboratory for the REACH analysis of rubber products. The two major machines required for REACH laboratory *viz.* GCMSMS and ICPMS were procured through GeM. Installation of the instruments is in progress.

6. Hands-on training

- Hands-on training for the manufacture of Exercise Band to M/s J K Polymers, Punjab.
- One day hands on training to Manufacture of carpet backing for M/s Oswal Polymers, Kottayam.
- One day specialized training to M/s. NHMP Multy Production Industries, Tripura in the production of fluorescent rubber bands
- Hands-on training to M/s Periyar Rubber Band on orthodontic rubber band preparation, which complies with the FDA regulations.
- Specialized hands on training to M/s Lenora Gloves (P) Ltd.on protein detection in rubber products.
- Training given to Shri. Hydross T A, Kothamangalam, on Industrial gloves manufacturing.
- Training given to Smt. R S Thankachi, Trivandrum for making Rubber band
- Glove manufacturing and latex coating on gunny jute bag was given to M/s Nambyattukudi Agro Industries, Kaladi

7. Factory visits/industry meets

- Officers participated in the seminar on start-up programme by Travancore Management Association
- An online talk about "Rubber products manufacturing in Kerala: challenges and opportunities" was conducted in the webinar hosted by MSME, Trissur on 23.07.2020.
- Attended the webinars "Basic approach to manage risk in mechanical testing laboratories" on 28/07/2020.
- Attended the webinars on ICPMS and GCMS hosted by M/s. Agilent and M/s. Thermofisher on 22/07/20 and 30/7/20 respectively.
- Attended webinar on "Natural Rubber Gloves Manufacturing: Prospects and Concerns" conducted by RTI on Friday, Sep 11, 2020, 2-5 pm.
- Attended online technology demonstration: "Tyre recycling using ultra high pressure water" on Tuesday, November 17, 2020 from organized by C. Ganga & NMCG.
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Retreading" on 18.12. 2020 organized by RTI-IRI.

8. Industrial training/ demonstrations

- Given webinars on topics "Latex products manufacturing" and "Overview of the rubber sector and scope for entrepreneurship" as part of the Entrepreneurship Development in Rubber Sector in Kerala on 20th August 2020.
- Conducted a one-day workshop on rubber produt manufacturing for one batch of students at CFSC, Changanacherry.

VII. Guidance to Ph.D and Masters thesis

- Three students are conducting PhD programmes in the Division.
- Three students from M/s. Central Institute of Petrochemicals Engineering & Technology (CIPET), Department of Chemicals and Petrochemical, Kochi joined for project work.

ECONOMICS DIVISION

The broad research areas of the Division comprises of farm management, primary processing and marketing, rubber products manufacturing industry and foreign trade, and intercrops and by-products. In these areas inter- divisional collaborate projects are also undertaken for comprehensive understanding of the sector. Details of work done during the reporting period are given in the following section.

1. Tariff policies and external trade performance of India's rubber and rubber products under the Regional Trade Agreements (RTAs)

The major objectives of the study are to analyse the tariffs and tariff policies on rubber and rubber products of India under its RTAs , to analyse the impact of tariff policies under the RTAs of India on the import of major rubber and products, and to highlight the policy implications based on the results of the study for a sustainable rubber industry of India. Analysis of impacts of tariff liberalisation under the RTAs on raw materials of rubber (19 tariff lines) and intermediate rubber products (11 tariff lines) found that in the case of NR no tariff lines exhibited growth in import due to the tariff policies under the RTAs of India. For SR, the tariff policies of India under the RTAs causes growth in import of five tariff lines and for RR also the tariff concession given under the RTAs was a major factor for the growth in imports into India. The results of the analysis on intermediate rubber products shows that more than the tariff policy, the growth in the domestic economy of India is the prime reason for higher import growth of items under the intermediate rubber products into the country.

2. Cost of cultivation of rubber in major rubber producing states of India

The state-wise information on actual cost of cultivation/production of rubber incurred by the farmers is essential for formulation of various policy measures.

But such state-wise actual cost of cultivation data is currently not available with Rubber Board. Hence, the Executive Director of Rubber Board has directed to estimate actual cost of cultivation/production of natural rubber in Kerala, Tamil Nadu, Karnataka, Odisha, Assam, Tripura, Meghalaya and Nagaland. Primary data will be collected from rubber growers of the aforesaid States using well-structured questionnaires. Separate questionnaires were developed for collecting data from immature and mature rubber plantations. Data will be collected from randomly selected respondents using officials of extension department of Rubber Board. 1000 units each from mature and immature plantations will be coved under the study. Data collection was initiated closing the reporting period.

3. Socio-economic impact of natural rubber cultivation under the block planting scheme in Tripura

The first comprehensive study on the socio economic impact of NR cultivation under the block planting scheme of Tripura was completed and reported during the year 2009-10. A survey in the same region was again carried out with the objective of capturing the decadal changes in the socio-economic characteristics of the tribal rubber growers in Tripura due to rubber cultivation. A total of 421 rubber growers in Tripura belonging to nine Block Planting Units (BPUs) and two Group Processing Centres (GPCs) were surveyed. The BPUs surveyed are Rani, Rambabu Para, Khamberbari, Dariabagma, Kariyamura, PS Para, RS Para, Kamalasagar and Laxmandepha and the two GPUs are Bagma and Janmabhumi. Data compilation and analysis is progressing.

4. Adoption of recommended practices for rubber cultivation by smallholdings

The Rubber Board recommends various rubber cultivation practices for increasing productivity and reducing cost of production of rubber. Though adoption of technologies developed by Rubber Board like improved clones are widely adopted by farmers, technologies recommended for better farm management are not well received by farmers. Though the Rubber Board took concerted efforts to popularise these rubber production technologies through its extension network, adoption by the farmers were not encouraging. In this backdrop, a study has been undertaken by the Division to study the level of adoption of different rubber production technologies by the smallholders, to study farmers' perception on rubber production technologies and to study the factors influencing technology adoption by rubber growers. A well-structured questionnaire was prepared for collecting data. Data will be collected from 1000 randomly selected farmers from South, Central, North-central and North Kerala. Descriptive statistics, Adoption Index, Adoption quotient and Likert Scale will be used for interpreting the data. A pilot survey was conducted during the reporting period covering 130 respondents.

5. Trends in rubber consumption in the non-tyre sector: An exploratory analysis

A study of the trends in consumption of rubber by the non-tyre manufacturers was undertaken to analyse the trends in the composition of consumption of rubber in the selected products, to identify the techno-economic factors contributing to the changes

in the composition of NR consumption in the non-tyre sector, to analyse the role of imports in determining the domestic composition of consumption in the non-tyre sector and to highlight the policy implications of the results. Filed survey was initiated during the reporting period.

6. Estimation of commercial rubber timber yield in Kerala

The project aims to estimate the timber yield from rubber estates in the traditional region. Rubber tree data of over 90,000 trees were collected from public sector undertakings like the State Farming Corporation of Kerala (SFCK), Rehabilitation Plantation Limited (RPL) and Arasu Rubber Corporation. Clone wise, felling age wise timber yield would be estimated using the data. Girth of trees at 150 cm from the bud union and length till the first branching was collected from the estates. Collation of collected data was initiated during the reporting period.

7. Agrarian distress and livelihood strategies of small farmers: the case of natural rubber in Kerala, India

The major objectives were to study the livelihood strategies of small farmers of natural rubber in India, especially in Kerala, in the background of the crisis in NR plantation sector consequent to the consistent fall in NR price for more than one decade since 2011. This study is based on both primary and secondary data sources. Primary survey will be conducted from 300 marginal, small, and medium households in three villages, Taluks and Districts each in three regions in Kerala (South, Central and North). The villages were identified using the data base of Rubber Production Incentive Scheme (RPIS) of the Government of Kerala. Accordingly, 114 samples in Elamade village in Kottarakara Taluk of Kollam district in southern Kerala, 73 Samples in Kondoor village in Meenachil Taluk in Kottayam district in central Kerala, and 113 samples in Thimiri village in Thaliparamba taluk of Kannur district in northern regions have been identified survey. Questionnaire required for the survey has been prepared and the survey will be initiated soon.

8. De-institutionalisation and its implications on the cultivation of natural rubber in India

The study was undertaken to explore the implications of the withdrawal of institutional interventions introduced since the inception of NR cultivation in the country on the sustainability of NR sector in the country. Secondary data was used for the study and data processing is progressing.

9. Role of Micro-level Clusters at Grass Root Level: The Case of Rubber Producers Societies in the Development of Natural Rubber Cultivation in India

The major objectives of the study were to examine the role of Rubber Producers Societies (RPSs) in the Development of Natural Rubber cultivation in India. The regional office-wise data required were collected from the RP Department of Rubber Board. Data compilation and processing are progressing.

CENTRAL EXPERIMENT STATION, CHETHACKAL, KERALA

The Central Experiment Station, Chethackal established in 1966 is situated at a distance of about 56 km from Kottayam. The Station has a total land area of 254.76 ha. The Station meets the field trial needs of the Scientists of various disciplines like crop improvement, crop management, crop protection, crop physiology, latex harvest technology and meteorological observation. The Station works under A and B Divisions of almost equal area. The priority areas of experimentation at present are breeding for high yield and other beneficial secondary characters with special emphasis on disease and drought tolerance, evaluation of clones developed conventionally, nutrition studies, intercropping systems, reduction in cost of cultivation, low frequency tapping systems etc.

Total number of research trials under various Divisions are 97. A three part tree crown budded area with canopy from FX 516 is laid to study disease resistance mechanisms. The Eddy Covariance tower gives micro environmental data. The Station is also functioning as a centre for training in various aspects in farming operations of rubber plantation industry.

During the reporting period, the total crop realized was 97.60 MT. A total of 272 tapping days was possible in the year and 56 tappers were engaged for tapping. The total man-days engaged in this Station were 31352 days. The Station having 153 permanent workers is managed by the Officer-in-charge with 26 staff for office administration, farm management, dispensary, security and canteen. The dispensary attached to the Station caters to the medical needs of the workers. The total patients attended to during the period under report were 2052. A medical camp for Antigen Test of Covid-19 has been conducted at CES Dispensary. The total IEBR raised during the reporting period was Rs. 1,27,45,867/-.

A Rubber Demo Project has been initiated in this Station in March 2021 as a part of Farm Tourism programme. A novel homestead planting system with rubber and 5 tropical fruit species was laid in a field trial in an area of 2.5 ha. The objective is to maximize utilization of land and sunlight for increasing land productivity and net income and the details are as follows:

Table CES. 1. **Details of homestead planting system**

Sl. No.	Treatments
1.	Rubber + Mangoosteen (20' x 20')
2.	Rubber + Achachairu (25 ' x 20')
3.	Rubber + Vietnam super early jack fruit (10' x 10')
4.	Rubber + Abiu (20' x 15')
5.	Rubber + Longkong (20' x 15')

The project on intercropping coffee, ginger and turmeric in mature rubber plantations to generate additional income for rubber growers were initiated.

REGIONAL RESEARCH STATION, GUWAHATI, ASSAM

1. Crop improvement

1.1. On-farm evaluation of selected ortets of *Hevea*

In an attempt to evaluate performance of selected promising ortets under the agro-climatic conditions of Assam, five genotypes (3 primary ortet selections *viz.* RRSG 9, RRSG 3 and RRSG 1 and two clones *viz.* RRIM 600 and RRII 429) were planted in large scale in Morigaon district. Girth of ortets after 4 years of planting was at par with RRIM 600. However, mean girth of RRII 429 was high compared to that of RRIM 600 and other ortets.

Table Ghy. 1. **Girth at 4th year of planting**

Name of clones/ortets	Mean girth at 4th year of planting (cm)	Range of girth (cm)
RRIM 600	25.8	17.0 - 38.8
RRSG 1	26.1	18.9 - 35.5
RRSG 3	26.2	19.0 - 34.5
RRSG 9	24.9	16.0 - 35.1
RRII 429	29.2	17.8 - 42.1
CD (Pd≤0.05)	NS	

1.2. On farm evaluation of potential clones

For final evaluation of potential clones of NE under the agroclimate of Assam, budding of SCATC 88/13, RRII 429, RRII 417, RRIM 600 and RRII 208 was completed and polybag plants were raised. Planting will be done in next season. Area was cleared and field preparation was under progress.

2. Crop management

2.1. Soil fertility mapping in Assam, Arunachal Pradesh, West Bengal, Manipur, Mizoram and Nagaland

In an attempt on survey of rubber growing soils of North-East India for developing soil fertility assessment based online fertilizer recommendation system, a total of 484 soil samples were collected from Assam. Location coordinates of all the soil samples were recorded using GPS device along with data related to cultural practices of each location. Soil sample collection from West Bengal, Arunachal Pradesh and Manipur were already completed.

REGIONAL RESEARCH STATION, AGARTALA, TRIPURA

The Station continued its research activities in four major areas *viz.* i) evaluation of clones, ii) crop management, iii) latex harvesting and iv) ecosystem studies. Advisory services on discriminatory fertilization to growers and DRC analysis are being continued.

1. Crop improvement

The crop improvement programmes are being continued for development and evaluation of clones for this region. Eighty eight selected clones *viz.* 20 hybrids, 21 OP progenies and 47 half-sib progenies were under evaluation in six clonal and seedling nurseries.

From clonal nursery evaluation of selected OP progenies (2013), seven OP progenies were planted with three check clones (RRII 208, RRII 429 and RRIM 600). Among the OP progenies highest girth was observed for AGOP 08A/23 *i.e.*, 43.3 cm and

is significantly superior girth compared to check clones. Mean highest test tap yield for last three seasons was recorded for AGOP08/31 *i.e.*, 151.7 g t^{-1} 10 t^{-1} which is higher than all the check clones. In another clonal nursery of OP progenies trial (2014), 14 OP progenies planted along with three check clones (RRIM 600, RRII 429 and RRII 208) highest test tap yield for last three seasons was recorded for AGOP 08B/15 *i.e.*, 115.4 g t^{-1} 10 t^{-1}which is higher than all the check clones.

In clonal nursery trial of selected half-sib progenies (2014), 13 selected half-sib clones planted with 4 check clones *i.e.*, RRIM 600, RRII 105, RRII 429 and RRII 208 AGHS 09/62 showed maximum girth *i.e.*, 41.2 cm among the clones. Highest mean test tap yield for last three seasons was recorded for AGHS 09/207 *i.e.*, 120.9 g t^{-1} 10 t^{-1} which was lower than check clones. In another half-sib progenies trial (2015), test tap yield data showed that AGHS 10/108 and AGHS 10/428 recorded 114.9 and 112.1 g t^{-1} 10 t^{-1} respectively. The check clones, RRII 208 and RRIM 600 recorded the test tap yield of 70.9 and 73.3 g t^{-1} 10 t^{-1} respectively.

In clonal nursery evaluation of selected hybrids (2013), 10 selected hybrids were cloned and planted with check clone RRII 208, RRII 429 and RRIM 600. The mean highest test tap yield for last three seasons was recorded for AGHY07/11 *i.e.*, 157.1 g t^{-1} 10 t^{-1}which is higher than all the check clones. In another hybrids trial (2015), AGHY 09/20 recorded highest average test tap yield of 41.7 g t^{-1} 10 t^{-1} followed by check clones, RRII 429 (40.2 g t^{-1} 10 t^{-1}), RRIM 600 (41.7 g t^{-1} 10 t^{-1}) and RRII 208 (44.1g t^{-1} 10 t^{-1}).

In large scale clone trial (2015) consisting 15 clones (DD/AGR/6/16, DD/AGR/6/5, RRSA 114, RRSA 121, RRSA 585, RRSG 248, RRST 37, 98/38, 98/46, 99/1/24, 99/5/9, RRII 208, RRIM 600, RRSA 98 and RRSA 315), clone DD/AGR 6/5 showed highest girth (47.8 cm) followed by DD/AGR/6/5 (42.0 cm) during sixth year compared to control RRIM 600 (39.8 cm).

In Genotype X Environment interaction trial (2019), the girth ranged from 3 cm to 7.45 cm and height ranged from 60 cm to 307 cm during second year among 57 genotypes. Higher girth (at 30 cm height) was recorded in P/77 (7.45 cm) followed by IAN 873 (7.26 cm) and RRII 208 (7.22 cm).

Standardization of distinctness, uniformity and stability (DUS) testing norms in 57 clones, 213 wild *Hevea* germplasm accessions and a breeding orchard are being maintained in the Station.

2. Crop management

The trial on evaluation of multi-faceted land use system under rubber based homestead farming is being continued. A popular fruit orchard has been established with seven species *viz.* Mango (*Mangifera indica*), Litchi (*Litchi chinensis*) Jackfruit (*Artocarpus heterophyllus*), Indian black berry (*Eugenia jambolan*) , Elephant apple (*Dillenia indica*), Pomelo (*Citrus grandis*) and Star gooseberry (*Phyllanthus acidus*) near homestead farming trial.

Evaluation of medicinal plants under mature and immature rubber (rectangular spacing) has been initiated at Taranagar farm. Six different species *viz.* Arjuna (*Terminalia arjuna*), Bakul (*Mimusope elengi*), Agar (*Aquilaria agallocha*), Amla (*Plyllanthus emblica*), Behera (*Terminalia bellirica*), Haritaki (*Terminalia chebula*) have been planted in inter row spaces available in the main crop of rubber plantation.

In evaluation of cropping system model, the girth of rubber in the intercropping plots (64.3 cm 64.1 cm respectively for Model I and II) and in monocropped plots (63.8 cm and 64.1 cm respectively for Model I and II) is statistically on par after 12 years of planting. The dry rubber yield obtained from intercrop and monocrop in both the Models are also on par *i.e.*, 1602 and 1578 kg ha^{-1} for intercrop and monocrop in Model I and 1542 kg ha^{-1} and 1576 kg ha^{-1} in Model II respectively.

In the skipping fertilizer experiment, the girth of rubber in 'No fertilizer plot' and conventional fertilized plots is statistically on par in both the years *i.e.* 57.1 and 57.7 cm during 2020-21 respectively. The mean yield in 'No fertilizer plot' and 'Conventional fertilized plots' was 1401 kg ha^{-1} and 1385 kg ha^{-1} during 2020-21 which is also statistically on par.

For zero tillage experiment the effect of different dimensions of pit on the growth and development of rubber plants was found to be statistically not significant at the end of 7th year *i.e* 50.3 for recommended pit size, 49.7 for poly bag size pits and 49.3 for root trainer size pits. Where the soils are deep and devoid of hardpans, irrespective of the size of the pits, the trees developed a long tap root of length 2.8 m, 2.9 m and 2.7 m for conventional pit size, poly bag pit size and root trainer pit size respectively after seven years after planting.

Goatery

A goatery unit has been started as a component of rubber based homestead farming system. This experiment was initiated with four black bengal goat (*Capra hircus*) which is a regional breed popular in Tripura state. Goat number has increased to seven due to addition of three kids during eight month period. The average birth weight of kid was one kg. The animals were vaccinated. They are allowed to graze during day time and supplement was fed during evening. Average weight of parent goats increased from 8.9 kg to12.8 kg with a monthly weight gain of 0.49 kg.

3. Latex harvesting study

Low frequency tapping in clone PB 235 with three system of tapping with stimulation was concluded. The results showed that S/2 d3 6d7 system of tapping with stimulation has recorded high yield compared to S/2 d4 6d7 or S/2 d7 6d7 systems of tapping. Significant difference was observed in dry rubber yield (g t^{-1} t^{-1}) among the treatments in all the years. The highest per tap yield was in d7 and lowest per tap yield was in d3 tapping system. Five year mean yield per tree was 80.8, 85.2 and 111.5 g t^{-1} t^{-1} in d3, d4 and d7 respectively. Five year mean annual yield was 2652, 2475 and 1873 kg/400 trees /year in d3, d4 and d7 respectively. Average number of tapping days in a year was 82 for d3 system, 65 for d4 system and 42 for d7 system of tapping.

In block trial experiment with RRIM 600 with stimulation in S/2 d3 6d7 system and S/2 d7 6d7 was concluded. The results showed that higher yield was recorded in d3 system compared to d7 system of tapping. Average three year dry rubber yield was 1312 kg /400 trees /year for d3 system and 1142 kg /400 trees /year for d7 system of tapping.

The experiment on effect of planting density on d3 tapping systems is being continued with clone RRII 429. Higher

yield per tree was observed in low density planting compared to high density.

4. Ecosystem study

A global climate model (GCM), BCC-CSM was used to construct the ecological niche distribution of *Hevea* species in North-East (NE) using four Representative Climate Pathways (RCP) *i.e.*, RPC 2.6, 4.5, 6.0 and 8.5. In the initial attempt, there was no significant difference between the GCMs and among the RCPs in terms of distribution pattern except in few pockets in Assam under present climate situations. The annual average temperature in 2000 climate scenario ranged from -9.0 to 26.1°C. The projected annual temperature would be varying from -6.0 to 28.2°C in 2050 for the same region indicating the climate being warmer in NE. The minimum temperature in winter season in NE is predicted to increase from 14.8°C in 2000 to 17.3°C in 2050. On the other hand, precipitation of warmest quarter (bio18) and coldest quarter (bio19) were contributing maximum in Western Ghats region under RCP 2.6, the most stringent scenario, whereas a different set of bio-climate *i.e.*, mean diurnal range of temperature (bio2), temperature annual range (bio7) and temperature seasonality (bio4) are important in rest of RCPs.

RCP studies in BCC-CSM climate model indicated that projected *Hevea* distribution with highest probability (>0.8) will be 7123 ha in NE under RCP 2.6, where expected increase in temperature ranges from 0.3 to 1.7°C. An estimated area of 8757 ha with probability of 0.6-0.8 of *Hevea* species presence would be available by 2050 in NE under RCP 4.5, where range of temperature increase is 1.1-2.6°C. Under the climate scenario of RCP 6.0, the potential distribution of *Hevea* species in NE will be around 5583 ha with Maxent probability of >0.8. Under this climate scenario, the average projected increase of ambient temperature is 0.8°C during 2046-2065. Under the nightmare scenarios of RCP 8.5, where average projected increase in temperature is 2°C with radiative forcing of 8.5 Wm^{-2}, the potential distribution of *Hevea* with highest suitability of more than 0.8 is 6161 ha.

The potential distribution of *Hevea* species in Western Ghats region is 6282 ha with maximum suitability/probability of more than 0.8 under RCP 2.6 of BCC-CSM global climate model. This maximally suitable area of 6282 ha is decreased to 5840 ha under RCP 6.0 which is 2.18 per cent of total area. However, the potential area under RCP 8.5 scenario is found to be 6439 ha, which is 2.4 per cent of the total area.

5. Advisory work

Total 64 soil samples from small holdings were analyzed and offered site specific fertilizer recommendation. Total 281 latex samples were tested for dry rubber content.

REGIONAL RESEARCH STATION, TURA, MEGHALAYA

The Regional Research Station, Tura continued its research activities on evaluation of clones, polyclonal population, crop physiology/ latex harvest technology and crop management.

1. Crop improvement

1.1. Poly-cross progeny evaluations

In the 2008 poly-cross progeny evaluation trial, a new set of Clonal Nursery Evaluation Trial-2014 has been started at the Rubber Board campus, Dakopgre, Tura in two designs in RBD. Plants are maintained in the field. In another set of 2011 populations, a total of 34 top yielders were selected on the basis of growth performance and juvenile yield and maintained in the field for further evaluation.

1.2. Half-sib progeny evaluation trial-2008 and 2009

Selected populations for both the years on the basis of growth performance and juvenile yield and are maintained in the field for further evaluation.

1.3 On-farm evaluation of selected clones

Three on farm trials were started during 2009 and 2010. Trial I includes blocks of six clones, *viz*. RRII 417, RRII 422, RRII 429, PB 235, RRII 203 and RRIM 600, in Mendipathar (North Garo Hills) and Trial II includes four clones *viz*. RRII 417, RRII 422, RRII 429 and RRIM 600 in Bolchugre, West Garo Hills. In the North Garo Hills, three years mean yield data were recorded in both the trials and preliminary data on yield states that highest mean annual yield was recorded in RRII 429 (39.6 g t^{-1} t^{-1}) closely followed by RRIM 600 (38.4 g t^{-1} t^{-1}), RRII 422 (35.2 g t^{-1} t^{-1}), PB 235 (34.4 g t^{-1} t^{-1}) and RRII 417 (30.8 g t^{-1} t^{-1}) and minimum yield was recorded in RRII 203 (25.4 g t^{-1} t^{-1}).

1.4. Evaluation of poly-cross progenies from four stations of NE region

The promising seedlings were screened on the basis of test tap yield among polyclonal seeds which were collected from four locations in the NE region *viz*. RES Nagrakata, RRS Agartala, RRS Guwahati and RRS Tura and the selected bud woods are maintained in the nursery for further study.

1.5. Nursery evaluation of poly-clonal seedlings trial 2013 and 2014

The poly clonal seeds collected from Poly Clonal Seed Garden, Mizoram were planted in the field in the year, 2013 at two locations of the RRS, Ganolgre farm and one location at R.B. campus, Dakopgre, Tura. On the basis of test tap yield, top 25 best performing progenies have been selected and maintained as bud-woods at Rubber Board campus, Tura and five progenies have been selected at RRS, Ganolgre farm for further evaluation.

1.6. Germplasm Arboretum at Teksragre farm

In order to maintain the 1st, 2nd, 3rd, 4th and 5th set of Germplasm Arboretum under the agro-climatic condition of Garo Hills of Meghalaya at Teksragre farm near Anogre, all the 604 accessions including Wickhams for 5 sets of Germplasm were planted in the field and maintained the plants. Field preparation work for 6th set of Germplasm planting is in progress.

2. Crop physiology/Latex harvesting technology

2.1. Effect of low winter temperature on yield of rubber at high altitude

Severe low winter temperature is one of the main factors for depression of yield and per cent dry rubber content in *Hevea* under the agro-climatic condition of Garo Hills. The annual mean yield (29.8 g t^{-1} t^{-1}) and DRC (34.0 %) for the reporting year was recorded. Low temperature adversely affected the yield and DRC. Early defoliation and refoliation was observed and during winter and DRC ranged observed from 28.8- 29.4 per cent. Lowest soil moisture content was also recorded in the months of February and March.

2.2. Shallow tapping – an option to stress alleviation in *Hevea* plantations during winter in NE

There was no significant difference between treatments. Maximum annual mean yield was recorded in normal tapping system (29.1 g t^{-1} t^{-1}) followed by normal continuous tapping (28.2 g t^{-1} t^{-1}) and LFT + normal tapping (27.9 g t^{-1} t^{-1}) and lowest was in shallow + normal tapping system (25.4 g t^{-1} t^{-1}). Annual DRC mean was low (33.7 %) in normal continuous tapping and high (34.2 %) in shallow + normal tapping system. Normal continuous tapping system showed higher TPD (7.0 %) followed by the shallow + normal tapping system (6.5 %) and LFT + normal tapping (6.2 %) and minimum was in normal tapping system (5.4 %).

2.3. Location specific stimulant application

Ethylene induced stress response in the tapping panel of the *Hevea* trees was initiated with the aim to reduce the stress in tissues in the tapping panel. In RRIM 600 six treatments were adopted with bark applications of five per cent Ethephon. Results showed that maximum annual mean yield (39.0 g t^{-1} t^{-1}) and low DRC (33.4 %) were recorded in T3 (Bark application of five per cent ethaphon at 150 cm above the bud union and near bud union and minimum yield (30.7 g t^{-1} t^{-1}) and high DRC (34.2 %) were recorded in T6 (Unstimulant trees). There was no significant difference in DRC between the treatments.

3. Crop management

3.1. Soil moisture retention characteristics under the rubber growing areas of Meghalaya

Soil samples were collected each month at the depth of 0-15 cm, 15-30 cm

Table Tura.1. **Rubber growing areas in each districts and the number of soil samples collected from each district of Meghalaya**

Districts	Area (Ha)			Number of soil samples
	Mature	Immature	Total	
North Garo Hills (NGH)	1304.2	2249.1	3557.3	55
West Garo Hills (WGH)	2394.7	4205.7	6600.3	162
East Garo Hills (EGH)	599.3	2115.1	2714.4	57
South Garo Hills (SGH)	495.6	1074.7	1570.2	39
South West Garo Hills (SWGH)	479.2	415.6	894.8	59
Ribhoi (East & West Khasi Hills)	119.5	950.5	1070.0	32
Total	5396.4	11010.6	16407.0	404

and 30-60 cm for soil moisture study. Soil moisture content increased with increase in the depth. Maximum was in August/ September and minimum was in Jan/Feb. Annual mean was 24.58, 25.38 and 26.16 %, respectively.

3.2. Analytical/advisory work for fertilizer recommendation

During the year, 24 soil samples collected from the rubber growing areas indicated that the O.C. content was at the medium range (0.81 to 1.32 %) in the surface soil (0-30 cm), available phosphorus was at low range (3.5 to 7.1 mg kg^{-1}) and available potassium was at medium range (75 to 96.1 mg kg^{-1}). The soil is acidic in nature with pH ranging from 4.6-5.5; fertilizer recommendation given accordingly.

3.3. Evaluation of soil fertility status and mapping in Meghalaya - A collaborative research project with Crop management group of RRII, Kottayam

Initiated the new project and collected 404 composite soil samples (0-30 cm depth) from the rubber growing areas of Garo hills of Meghalaya (West, East, North, South, South-west Garo Hills districts and Ribhoi/ East and Khasi hills district) using GPS system with the help of Rubber Board, Regional Office Tura Field officers, Sri. Babu P, Jr. Farm officer, RRII, Kottayam and Dr. Debasis Mandal, Senior Scientist, RRS, Agartala . All the 404 soil samples collected from Garo Hills and Ribhoi/ East and Khasi Hills of Meghalaya were transported to RRII, Kottayam for analytical and fertility mapping works for RUBSIS study.

Four hundred and four soil samples (North Garo Hills n=55, West Garo Hill n= 162, East Garo Hills n=57, South Garo Hills n=39, South West Garo Hills n=59 and Ribhoi/East and West Khasi Hills n=32) were collected from the rubber growing areas of Meghalaya under the North-eastern states and analyzed for available macro-nutrients and micro-nutrients status of 13 parameters. One composite soil sample (0-30 cm) was collected for each 30 ha rubber areas. Soil samples were collected from Blocks with more than five per cent area under rubber. Samples were collected both from mature and immature areas in the proportion of 1:2, considering that approximately 67 percent of the total rubber is in immature phase (Table Tura 1). Soil samples were collected during December, 2017 to November, 2019. Global Positioning System (GPS Garmin Dakota 20) devices were used for recording location of soil samples. Spatial distribution map of rubber area was overlaid over soil samples of each district in GIS environment for checking the distribution pattern and revisits were made whenever necessary to ensure adequate representation of the entire rubber area. Soils varied from sandy loam to clay loam in texture, extreme to moderately acidic in reaction (pH: 4.0 to 6.3), medium to high organic carbon content (8.3 to 34.1 g kg^{-1}), available phosphorus was very low (0.1 to 327.2 mg kg^{-1}), available potassium varied from low to medium (10.6 to 727.9 mg kg^{-1}) (Table Tura 2a). Available calcium (12.3 to 3000.7 mg kg^{-1}) and magnesium (8.0 to 2227.0 mg kg^{-1}) showed high in fertility ranged and available sulphur showed medium range (0.0 to 158.4 mg kg^{-1}) 9. DTPA extractable B, Cu, Mn, Fe, Zn and exchangeable Al content of of the soil samples of Meghalaya ranged from 0.04 to 2.7 (mean 0.6 mg kg^{-1}), 0.0 to 5.2 (mean 1.04 mg kg^{-1}), 4.0 to 391.5 (mean 60.0 mg kg^{-1}), 5.7 to 8.0 (45.7 mg kg^{-1}), 0.1 to 74.6 (mean 3.8 mg kg^{-1}) and 0.0 to 4.6 (mean 1.2 cmol (P+) kg^{-1}) of soil, respectively.

Table Tura. 2. **Descriptive statistics of available macro-nutrients (Primary) in rubber growing soils of Meghalaya under N.E. India**

	Soil pH					Organ Carbon g kg^{-1}					Phosphorus (P) mg kg^{-1}					Potassium (K) mg kg^{-1}				
	Min	Max	Mean	SD	CV	Min	Max	Mean	SD	CV	Min	Max	Mean	SD	CV	Min	Max	Mean	SD	CV
Meghalaya																				
North Garo Hills (NGH)	4.1	5.3	4.7	0.3	5.6	8.6	26.4	16.6	0.4	26.4	0.82	8.93	3.7	2.1	56.3	10.6	299.9	150.5	65.9	43.8
West Garo Hills (WGH)	4.1	6.1	4.6	0.3	6.4	5.6	40.4	16.9	0.6	34.6	0.1	327.22	14.6	43.9	293.3	19.4	367.7	104.4	60.0	57.5
East Garo Hills (EGH)	4.0	5.4	4.7	0.2	5.0	11.8	36.7	24	0.6	23.6	1.3	15.47	3.9	2.5	64.5	40.6	727.9	142.8	116.3	81.4
South Garo Hills (SGH)	4.1	5.2	4.6	60.2	4.7	8.2	49.5	17.5	0.8	5.6	2.2	266.81	7.3	55.0	317.3	29.2	87.6	54.0	14.6	27.0
South West Garo Hills (SWGH)	4.2	5.6	4.7	0.3	6.8	5.1	23.5	12.8	0.4	32.6	2.4	60.6	6.1	8.0	131.9	28.1	214.4	89.4	44.2	49.4
Ribhoi/East & West Khasi Hills	4.1	6.3	4.7	0.5	10.4	10.6	8.2	6.2	0.4	23.9	0.5	35.11	4.2	7.9	188.8	32.0	339.5	28.0	82.0	64.1
Mean of Meghalaya state	4.1	5.7	4.8	0.3	6.5	8.3	34.1	17.33	0.5	31.1	1.2	119.02	8.3	19.9	175.3	26.7	339.5	111.5	63.8	53.9

REGIONAL EXPERIMENT STATION, NAGRAKATA, WEST BENGAL

1. Crop improvement

1.1. Evaluation of clone

Twenty six promising clones were evaluated under the agro-climatic conditions of sub-Himalayan West Bengal. After 27th year of planting, significantly higher girth were exhibited by clones PR 107, SCATC 93/114, RRIM 703, RRIM 605, RRII 118 and RRIM 612 than check clone RRIM 600. Two clones PB 310 and PB 280 recorded significantly higher rubber yield than check clone RRIM 600.

1.2. Evaluation of germplasm

Germplasm evaluation trials were being continued at RES, Nagrakata comprising of 21 accessions. Highest growth was observed in RO 2629, MT 44, RO 3430, RO 2635, MT 196, MT 2229, AC 619, RO 5557 and RO 2890. As compared to check clone RRII 105, significantly higher dry rubber yield was recorded in AC 763.

1.3. Multi trait screening of half sib progenies for cold tolerance and yield attributes

Half-sib progenies were raised from seven different clones in 2014. The juvenile yield of progenies raised from seeds of SCATC 88-13 during non-winter period showed the highest yield followed by RO 5363. During winter period, the average juvenile yield of SCATC 88-13 progenies was higher than that of RRIM 600, followed by RRII 417 and RRII 429. Number of seedling plants showing above average juvenile yield was also higher in SCATC 88-13. The potential of half-sib progenies of SCATC 88-

Table Nag. 1. **Performance of trees showing above 50 g t^{-1} t^{-1} yield every year over four years**

Category	Mother plants	Girth at 12th year at 50 cm height (cm)	Mean yield over four years g t^{-1} t^{-1}	Pre-winter yield contribution (%)	Category	Mother plants	Girth at 12th year at 50 cm height (cm)	Mean yield over four years g t^{-1} t^{-1}	Pre-winter yield contribution (%)
A	WBGHY 112	69.2	65.3	39.6	N	WBNGK 146	79.2	79.7	42.4
	WBGHY 182	64.9	63.0	56.4		WBNGK 276	82.8	63.1	45.8
	WBGHY 248	85.4	86.7	41.7		WBNGK 297	81.0	56.2	51.8
	WBGHY 32	64.5	53.8	54.0		WBNGK 379	84.7	58.3	51.5
	WBGHY 335	64.3	67.5	39.3		Population mean	81.93	64.3	47.9
	WBGHY 340	89.0	56.9	55.5	T	WBTUR 11	75.8	56.2	55.5
	WBGHY 385	77.0	58.8	52.9		WBTUR 200	65.6	71.1	48.2
	WBGHY 390	72.7	67.6	45.4		WBTUR 213	86.2	53.0	56.0
	WBGHY 41	89.2	104.1	41.7		WBTUR 320	99.5	57.4	59.5
	Population mean	75.13	69.3	47.4		WBTUR 327	79.8	70.2	48.9
K	WBKAN 282	80.0	54.0	49.2		WBTUR 328	73.3	59.0	40.7
						WBTUR 69	85.7	66.5	50.4
						Population mean	80.8	61.9	51.3

13 was shown to be prominent from this study.

1.4. Performance of new generation clones under the agro-climate of sub-Himalayan West Bengal

Evaluation of five promising new generation clones under the cold agro-climate of sub-Himalayan West Bengal showed that girth and yield of all the clones were on par with RRIM 600. However, yield of RRII 422 was found high followed by RRII 429 and RRII 417 during the year. Comparatively RRII 414 recorded lower yield than other clones in this region.

2. Crop physiology

2.1. Performance of polycross progenies raised from seeds of locally adapted mature rubber plantation

Seeds were collected from Kamrup, Assam; Jalpaiguri, West Bengal; Tura, Meghalaya and Kanyakumari, Tamilnadu for the evaluation of seedling trees raised from seeds developed across diverse environment. Seedling trees were planted under the agroclimate of Sub-Himalayan West Bengal. Girth and yield of seedlings from varied seed sources was not significantly different over initial four years of tapping. Overall, 21 per cent seedling trees showed above 50 g $t^{-1}t^{-1}$ yield among all the tapping trees. Trees showing above average yield were high in seedling trees raised from seeds of Kanyakumari, Tamilnadu followed by that of Tura, Meghalaya. However, in case of seedlings showing above 50 g $t^{-1}t^{-1}$ yield, it was high in seedlings sourced from Guwahati (Table Nag. 1). All the total 21 seedling trees were screened on the basis of mature yield that showed above 50 g $t^{-1}t^{-1}$ yield every year over four years. Dominance of seedling trees from Guwahati seed-source showing

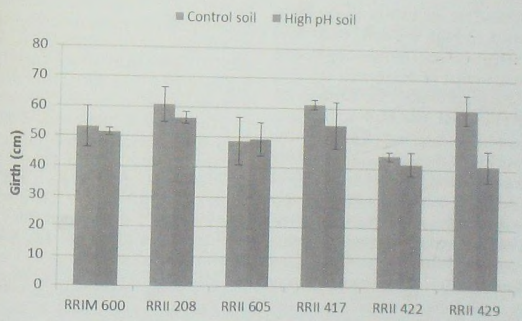

Fig. Nag. 1. Girth of rubber clones in control soil (pH 5.5) at RES, Nagrakata and high pH soil (pH 7.9) found in abandoned tea growing areas of Dooars belt of North Bengal

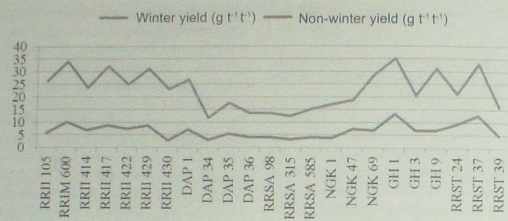

Fig. Nag. 2. The winter and non-winter yield of ortet selections and check clones at RES, Nagrakata

above 50 g $t^{-1}t^{-1}$ yield every year was noticed followed by that of Tura. Five seedling trees showed high girth and yield which were pinned for further studies to generate new planting material for NE India.

2.2. Physiological evaluation of rubber clones in abandoned tea growing areas of Dooars belt of North Bengal

A total of six clones were under evaluation in high pH soil. Growth of plants in high pH soil (7.9) was appreciable but lower to that of the normal soil (5.5) after eight years of planting. Girth at 125 cm height from the bud union was considered as a measure of growth. The growth of RRII 208 and RRII 417 was better in both control and high pH soil. RRII 422 and RRIM 605 recorded comparatively lesser growth after eight years (Fig. Nag. 1). RRII 429 showed good growth in control soil at par with RRII 208 and RRII 417 but in high pH soil RRII 429 showed very poor growth. Tapping has been initiated in September 2020.

2.3. Evaluation of Ortets for abiotic stress tolerance in different agro-climatic regions

Among the 23 ortet selections and popular check clones, RRII 429 showed significantly high girth after eight years of growth at RES, Nagrakata, which is a cold prone region. Among the ortet selections GH 1, GH 3 (selections from RRS, Guwahati) and RRST 37 (selection from RRS, Tura) showed higher girths than RRIM 600 the most popular clone of NE region. Ortet selections *viz.*, GH 1, GH 3, RRSA 98 and RRST 37 showed minimum defoliation during winter (wintering) close to RRII 429. RRIM 600 showed higher wintering percentage. The winter and non-winter yield of ortet selection GH 1 superseded the yield of popular clones RRIM 600 and RRII 429. Ortet selections RRST 37 and GH 9 were also found to be good yielders (Fig. Nag. 2).

REGIONAL RESEARCH STATION DAPCHARI, MAHARASHTRA

The thrust areas of research of the Station are environmental crop physiology and crop improvement and there are 15 on going trials in the Station in these two areas. There are two LSTs in environmental crop physiology and in crop improvement there are 2 LSTs, 3 FETs and 8 clonal nursery evaluation trials.

1. Evaluation of environmental stress tolerance and physiological adaptations of cold and drought tolerant ortet selections under varying agro-climates in India

The trial was laid out with RBD in July 2012 with 15 ortets from Agartala, Tura, Nagarkata, Dapchari and Guwahati with 7 check clones. Recording of causality counts, visual scoring of drought, growth parameters *etc.* were carried out. A significant difference in girth was recorded. Girth ranged from 19.7, 25.4 cm in Dap 36 to 34.2, 38.9 cm in RRSA 315 during 2020 and 2021. Among the ortets RRSA 315 recorded higher girth of 34.2 cm in March 2020 and continued to attain higher girth in March 2021 (38.9 cm) while in check clones, the highest girth was recorded in clone RRII 417 (33, 37.9 cm in 2020, 2021 respectively). In 2020 girth is at par for all Agartala ortets, GH 3, RRST 39 and all check clones except RRIM 600 while in 2021 all ortets from Agartala and Tura all check clones GH 3 and DAP 35 are at par. In general, ortet RRSA is superior in growth characters studied (31.7, 36 cm girth in March 2020 & March 2021). Tura had IInd rank, GH ortets had IIIrd rank and DAP ortets were poor performers under this agro-climatic condition.

2. Screening of wild *Hevea* accession for drought under Dapchari condition

The trial was laid out in July 2003 using 130 wild germplasm accession along with selected clones RRII 105, RRIM 600, Tjir 1 and RRII 208 as checks (control) in augmented RBD with plot size of five and a spacing of 2.5 x 2.5 m. Observation on growth parameters were recorded at pre drought period. The accessions showed wide variability for all characters studied. In general, Mato Grosso accessions were superior for all the growth characters studied than those from the Acre and Rondonia provenances. Among the control clones RRIM 600 and RRII 208 were superior to RRII 105. Twenty five potential drought tolerant accessions were identified based on 3-4 years field performance for further and detailed studies and are in progress.

3. Further field evaluation of selected *Hevea* clones for drought tolerance

The trial was laid out in July 2007 using 23 potential drought tolerant accessions along with five HP clones and selected clones RRII 105, RRII 430, RRIM 600, Tjir 1 and RRII 208 as check (control) in augmented RBD with plot size of five and a spacing of 2.5 x 2.5 m. Observation on growth parameters were recorded. The accessions showed wide variability for all characters studied. Among the control clones RRIM 600 and RRII 208 was superior to RRII 105.

4. Further evaluation of Dapchari ortets selected in LST

Trial started during 2008 to evaluate the growth and yield performance of ortets selected from polycross seedling planted at this station with control clones with the objective to evaluate further growth and yield potential of Dapchari ortets selections from polyclonal populations. Significant difference in girth was noticed. Among the check clones girth ranged from 37.4, 38.9 cm in RRII 105 to 41.9, 44.3 cm in RRII 430 in 2020 and 2021. Among the ortets OS 173 and OS 1 recorded higher girth of 43.9 and 47.1 cm in 2020, 2021 while lowest girth was noticed in ortet OS 236 (29.1, 26.6 cm) in 2020 and 2021 respectively. In 2020 girth was at par for all ortets except OS 8, OS 42, 136, 236, 317 and RRIM 600 while in 2021 all ortets and check clones were at par except OS 236. It was also noticed that all ortets were superior in girth to clone RRII 105 except ortet OS 42, OS 111, OS 317, OS 136, OS 236 in 2020 and 2021.

Table Dap. 1. **Girth in various ortets selected from Dapchari (2019-21)**

Clones	20-Mar	21-Mar	Clones	20-Mar	21-Mar
OS 1	43.7	47.1	OS 135	38.4	40.0
OS 8	36.0	39.2	OS 136	35.5	37.7
OS 34	40.7	43.1	OS 173	43.9	46.7
OS 35	42.4	45.6	OS 216	43.0	46.6
OS 36	42.8	45.3	OS 236	29.1	26.6
OS 37	39.5	41.8	OS 317	33.6	33.6
OS 42	33.4	35.7	RRII 105	37.4	38.9
OS 111	37.7	40.0	—	—	—

5. Small scale further field evaluation trial of selected wild accession for drought tolerance

Experiment was initiated during 2010 with 47 selection from wild *Hevea* accessions along with four check clones (RRII 105, RRII 208, RRII 430, RRIM 600) with the objective to confirm the drought tolerance potential of selected seven wild accessions from preliminary field screening by growing them at normal spacing at drought prone region and subjecting to detailed studies along with recording mature yield. The trial is in initial stage.

6. Clonal nursery evaluation of promising *Hevea* clones (Half sib progeny of prepotent clones) in hot spot areas for drought tolerance

A clonal nursery experiment with clones selected from half sib progeny of prepotent clone was initiated in 2010 with the objective to evaluate the clones in a clonal nursey and advance the potential ones showing drought tolerance along with rubber yield to LST and PCE to reduce the breeding cycle. The trial is in initial stage.

7. Field evaluation trial of selected wild accessions for drought

Trial based in rectangular lattice design was laid out in 2010 with 11 wild *Hevea* accessions along with two check clone (RRII 105 and RRIM 600) in order to evaluate juvenile and mature performance under drought condition. The trial is in initial stage. Recording of growth data and drought scoring will be carried out. The experiment is in initial stage.

8. Clonal nursery evaluation of pipe line clones for drought tolerance

Trial was laid out in July 2011 using 50 pipeline clones and two check clones in rectangular lattice design at spacing of 2.5 x 2.5 m with the objective to identify drought

tolerant clones for their adaptability and stability to the agro-climatic condition of Maharashtra. Observation on growth parameters were recorded. The test tapping was conducted for two years. Clone responses for field establishment were assessed. The 14 pipeline clones were found to be superior to clone RRII 430. Highest test tapping yield was recorded in P 20 along with RRII 430, while 11 clones performed better than clone RRII 105 and showed the local adaptive nature of pipeline clones.

9. Lager scale pipeline clones for drought tolerance

Trial was laid out in 2018 with the objective to evaluate growth and yield potential of pipeline clones and to select drought tolerant clones for this region. Growth and drought scoring and survival percentage was recorded. The trial was maintained and survival and adoptability assessment was being carried out. Check clones RRII 430 and RRIM 600 showed 12.9 and 13.4 per cent leaf drying while check clone RRII 105 showed more leaf drying (18.5%). Pipeline clones P 114 (3.7%), P 192 (5.7%), P 200 (6.8%), P 225 (8.4%) and P 68(8.9%) had considerably lesser leaf drying than RRII 430. Clones P 205 (17.8%), P 27 (18.3%), P 207 (21.4%) and P 196 (21.7%) showed the highest leaf drying among all the clone tested. Two top ranking clone in terms of leaf drying (P 114, P 192) also maintained relatively more number of whorls and leaves and these clones had more height indicating their better initial growth in the field in a drought prone region.

10. Development of drought tolerant root stock for the non traditional areas (2015)

Trial laid out in 2015 aimed at developing drought tolerant root stocks for the non traditional area by evaluating the drought tolerance capacity of the seedlings produced from seeds of drought tolerant clones and seeds from trees grown in drought prone non traditional areas as against the seedlings from traditional areas. The root stocks showed a wide variability for all characters studied. Out of the 40 clones and nine controls planted in this trial, 16 clones in terms of growth and 25 clones in terms of test tap yield were found superior to the check clones RRII 422 and RRII 417. The selection 69 showed a superior growth and yield performance.

11. Clonal nursery evaluation of selected progenies of the cross RRII 105 X PB 280 and its reciprocal for drought tolerance

The trial aims to identify drought tolerance capacity of the selected progenies of the 1996 HP, by evaluating them in small scale clonal nursery in drought prone area and to select drought tolerant clones for the non traditional area.Trial is in initial stage.

12. Evaluation and selection from progenies of polyclonal seed gardens and multi-clone populations

Selection of progenies of polyclonal seed garden and multiclone populations from different locations was made and tested in different agro climate. The evaluation resulted in the selection of 146 superior progenies based on test tap yield and girth. Higher number of selection was obtained from progenies from Tura. The trial was completed and multiplications of the selections for nursery was done.

13. Clonal nursery evaluation of pipeline clones/ marker assisted selection for drought and cold tolerance

The trial aimed to evaluate the drought tolerance potentiality of pipeline clones/ marker assisted selections under drought conditions and to select drought tolerant pipeline / marker assisted clones for this region. Survival percentage was recorded. The trial is in initial stage.

14. Evaluation of polyclones of Kanyakumari origin in clonal nursery

The trial aimed to study the growth and yield performance of polyclonal seedlings at early stage in the nursery and to examine the scope for the early selection based on dependable juvenile traits under rainfed condition. The trial is in initial stage.

REGIONAL RESEARCH STATION, DHENKANAL, ODISHA

The Station continued its research activities with the particular objective of screening best clone for planting and to develop location specific clones, suited to the dry sub humid climate region and the Odisha state and to provide technical guidance to rubber growers for rubber farming and processing aspects in the region.

1. Crop improvement

The clone evaluation trials are in progress with the prime aim to screen and evolve the most suitable and high yielding and adaptable location specific clone for drought prone region and for Odisha state.

1.1. Clone evaluation

In Trial 1 (1987), the elite clone RRIM 600 has recorded highest mean yield of 57.1 g $t^{-1}t^{-1}$ and GT 1 recorded the lowest yield (45.0 g $t^{-1}t^{-1}$). RRIM 600 is the preferred clone in the region. Further, GT 1 (87.9 cm) has recorded significantly higher mean girth (87.9 cm) over RRII 105 and RRIM 600. In terms of growth all three clones recorded good performance and also showed good adaptability in the region (Table OD. 1).

Table OD.1. **Yield and growth performance of elite clones**

Clone	Yield (g $t^{-1}t^{-1}t^{-1}$)	Girth (cm)
RRII 105	47.6	79.4
RRIM 600	57.1	83.6
GT 1	45.0	87.9
CD(P=0.05)	8.02	4.88

In clone trial 1990, RRII 208 (63.5 g $t^{-1}t^{-1}$), SCATC 88-13 (75.3 g $t^{-1}t^{-1}$) and RRIM 600 (63.4 g $t^{-1}t^{-1}$) found most high yielding clones. RRII 208 way found most promising clones in the region in terms of both yield and growth and adaptability. Other popular clones also performed well in the region. SCATC 93-14 recorded comparatively lower yield (36.0 g $t^{-1}t^{-1}$). However SCATC 93-14

recorded comparatively best growth in terms of girth, followed by SCATC 88-13 and RRII 208 (Table OD. 2).

Table OD. 2. **Growth and yield performance of differentclones in Odisha**

Clones	Yield ($g^{-1}t^{-1}t^{1}$)	Girth (cm)
Haiken 1	46.1	89.8
RRIM 600	63.4	90.7
RRIM 701	49.5	93.1
RRII 5	52.4	92.5
SCATC 88-13	75.3	100.0
SCATC 93-14	36.0	100.3
PB 310	45.6	93.2
RRII 208	62.5	92.7
PCK 1	50.5	93.1
RRII 300	54.3	93.2
C.D. (P=0.05)	12.46	–

In the 1991 clone evaluation trial, clones differed significantly in mean yield production. RRII 208 (75.5 g $t^{-1}t^{-1}$), RRIM 600 (63.4 g $t^{-1}t^{-1}$) and RRIC 102 recorded highest yield among the clones. RRII 208 was found high yielding and most suitable clone for Odisha region. Polyclonal seedling population (44.0 g $t^{-1}t^{-1}$) yielded low though having better growth and adaptability under the prevailed stress conditions (Table OD. 3).

In the other modern clones' trial (2000), the highest mean yield was observed in RRII 208 (60.1 g $t^{-1}t^{-1}$) and IRCA 109 (57.1 g $t^{-1}t^{-1}$). RRII 208 was found most promising clone in the region. IRCA series clones also performed well in the region. The lowest mean yield was recorded in RRII 51 (39.7 g $t^{-1}t^{-1}$). Highest growth in terms of girth was observed in RRII 300 (81.9 cm) (Table OD. 4).

Table OD. 3. **Performance of different clones in comparison to poly clonal seedlings in the Odisha region**

Clones	Yield ($g^{-1}t^{-1}t^{-1}$)	Girth (cm)
GT 1	48.8	105.9
RRII 105	64.1	91.3
RRII 208	75.5	102.3
RRII 5	54.5	94.9
RRII 300	54.4	101.0
PR 261	56.0	92.5
PR 255	58.5	102.6
RRIC 102	66.1	95.3
RRIM 600	63.4	91.1
Polyclonal	44.0	117.9
C.D. (P= 0.05)	14.57	9.65

Table OD. 4. **Yield and growth performance of modern clones**

Clones	Yield ($g^{-1}t^{-1}t^{-1}$)	Girth (cm)
RRII 300	46.7	81.9
RRII 208	60.1	64.0
RRII 357	44.0	64.0
RRII 352	51.1	63.1
PB 28/59	51.7	65.7
RRIM 600	59.5	67.5
RRII 351	51.1	61.8
IRCA 109	57.1	51.6
RRII 105	56.2	59.2
RRII 51	39.7	63.1
IRCA 111	53.6	66.5
C.D.(P=0.05)	9.88	10.32

REGIONAL RESEARCH STATION, PADIYOOR, KERALA

Identification and evaluation of clones adaptable for commercial cultivation in the region, development of agro management techniques for improved production/productivity with reduction in gestation period and clonal tolerance to drought/disease are the major thrust areas of research in the Station. The Station has a well maintained agromet observatory and a source bush nursery of promising clones and ortet selections suited to the region.

Table PAD. 1. **Ongoing trials**

Sl No	Experiment	Year of commencement
	CROP IMPROVEMENT	
1	Investigations on Genotype X Environment interaction in *Hevea brasiliensis*	1996
2	Clone evaluation trial	1996
3	Large scale trial of potential hybrid clones 1989 SST selections	1996
4	Large scale trial of potential hybrid clones 1996 SST selections	1996
5	Participatory clone evaluation- O F T	2019
	CROP MANAGEMENT	
6	Response to applied fertilizers in high yielding clones of *Hevea*	2002
	CROP PROTECTION	
7	Disease evaluation in clones of *Hevea*	1997
	CROP IMPROVEMENT, GERMPLASM	
8	Screening of *Hevea* germplasm for latex timber traits	2000
9	Preliminary evaluation trial 2000 A	2000
10	Preliminary evaluation trial 2000 B	2000
11	Clonal nursery evaluation of half-sibs	2009
12	HP programmes with domesticated and wild accessions	2000

CONCLUDED TRIALS

Sl. No	Experiment	Project Leader
1	Further evaluation and selection of wild *Hevea* germplasm (FET 1995)	C.P. Reghu
2	Physico chemical characterization of soils at the RRS, Padiyoor in the North Malabar region of Kerala	Radha Lakshmanan
3	Effect of irrigation on growth and water use efficiency of *Hevea*	Radha Lakshmanan
4	Clonal nursery evaluation of polycross progenies	

	for drought tolerance in North Malabar	Kavitha K. Mydin
5	Preliminary evaluation trial 2002	G.P. Rao
6	Evaluation of *Hevea* clones and ortet selections at high altitude situations, Ambalavayal	Radha Lakshmanan
7	Water consumption in rubber nurseries	Radha Lakshmanan
8	Mechanised land preparation for *Hevea* planting and plant growth	Radha Lakshmanan

The 24 tapping blocks in the farm was converted from the 1/2S d3 system of tapping to weekly tapping (1/2S d6 6d/7) from 2020 June onwards.

HEVEA BREEDING SUB-STATION KADABA, KARNATAKA

Hevea Breeding Sub-station (HBSS) with a research farm at Nettana was established in 1986. The major constraints in commercial cultivation in this region is drought in summer months and occurrence *Phytophthora* and *Corynespora* leaf fall diseases. The research programmes in the Station are envisaged to identify clones tolerant to different biotic and abiotic stress factors and to identify locally adapted clones for South Konkan region. The farm has a source bush nursery of 106 clones for generating nucleus planting material and a well-established Class B Agro-meteorological Observatory. The five ongoing trials in the station are as follows:

1. Large scale clone evaluation trial (1990)
2. Small scale evaluation trial 1991 A
3. Small scale evaluation trial 1991 B
4. Small scale evaluation trial 1991 C
5. Large scale evaluation trial 2000

1. Large scale clone evaluation trial (1990)

In LST 1990 trial HP 372 recorded highest yield than other clones but is on par with HP 223. All other clones recorded comparable yield with each other. The lowest yield was registered by Tjir 1 (45.2 g $^{-1}t^{-1}t^{-1}$) (Table Kad. 1).

Table Kad. 1. **Yield from large scale clone evaluation trial (1990)**

Sl. No.	Clones	Yield ($g\,t^{-1}\,t^{-1}$)	Sl. No.	Clones	Yield ($g\,t^{-1}\,t^{-1}$)
1	PB 260	84.2	9	PB 311	64.5
2	HP 223	89.4	10	Tjir 1	45.2
3	Mil 3/2	66.9	11	Gl 1	45.3
4	HP 204	55.7	12	Hil 28	55.4
5	HP 185	64.3	13	GT 1	58.7
6	PB 217	68.9	14	PB 235	78.2
7	RRII 105	55.8	15	HP 187	60.5
8	HP 372	95.6	—	—	—
	SE.d	7.85		CD (p=0.05)	16.4

2. Small scale clone evaluation trial (1991 A)

In 1991, three small scale trials were planted *viz.*, 1991 A (36 clones), 1991 B (13 clones) and 1991 C (13 cones). In the trial 1991 A with 36 clones, PB 235 recorded

higher yield (59.9 $g^{-1}t^{-1}t^{-1}$) which was followed by RRII 6 and were significantly higher than RRII 105. The lowest yield was recorded by RRII 105 followed by AVROS 352 and all other clones were recorded comparable yield with each other (Table Kad. 2).

Table Kad. 2. **Cup lump yield from small scale clone evaluation trial**

Clones	Cup lump yield ($g\,t^{-1}\,t^{-1}$)	Clones	Cup lump yield ($g\,t^{-1}\,t^{-1}$)
RRIC 36	15.7	PB 280	24.7
PB 314	36.5	PB 255	20.0
SCATC 88	15.1	RRIM 703	15.2
PB 312	21.0	PB 310	36.9
RRIC 104	13.9	PB 217	23.7
RRIC 100	22.4	RRIM 701	13.7
O 63	18.1	PB 5/60	17.5
KRS 163	13.5	WARRING 4	11.3
LCB 1320	16.0	RRIM 605	17.2
AVROS 352	10.4	CH 4	12.1
P 46	16.7	CH 26	29.7
RRIM 501	19.2	RRII 6	46.4
PB 235	59.9	RRIM 600	12.1
RRII 105	10.3	KRS 25	11.2
RRII 203	29.4	SCATC 93-11	11.5
PB 260	17.9	HAIKEN 1	25.0
RRII 300	46.4	GT 1	13.5
PB 311	27.8	KRS 128	13.1
SE.d	3.5	CD (p=0.05)	7.2

3. Small scale clone evaluation trial (1991 B)

Among the 13 clones evaluated in the trial 1991 B, RRII 118 (37.5 $g\,t^{-1}t^{-1}$) recorded highest mean yield and was significantly higher than RRII 105 (Table Kad. 3).

4. Small scale clone evaluation trial (1991 C)

No significant different was observed among the clones with respect to yield. But among the clones evaluated PR 261 recorded numerically higher yield than the other clones.

Table Kad. 3. **Yield from small scale clone evaluation trial (1991 B)**

Clones	Yield ($g\,t^{-1}\,t^{-1}$)	Clones	Yield ($g\,t^{-1}\,t^{-1}$)
RRII 208	7.0	RRIC 102	8.4
CH 3	8.2	RRII 5	39.1
RRII 3	24.0	AVROS 255	19.6
CH 2	11.5	PB 5/139	2.3
GT 1	25.0	NAB 17	16.5
RRII 118	37.5	RRII 105	8.7
RRII 308	17.8	—	—
SE.d	2.51	CD (p=0.05)	6.57

5. Large scale trial (2000)

The large scale trial for evaluation planted in the year 2000 consists of hybrids RRII 403, RRII 407, RRII 414, RRII 422, RRII 429 and RRII 430 and their parents *viz.*,RRIC 100 and RRII 105. Significant difference in yield was observed between the clones tested and the highest mean yield was recorded for the clone RRII 414 (86.7 $g\,t^{-1}t^{-1}$) which is on par with clone RRII 430. Check clone RRII 105 recorded 41.1 $g\,t^{-1}t^{-1}$ (Table Kad. 4).

Table Kar. 4. **Yield from large scale clone evaluation trial (2000)**

Sl. No.	Clone	Yield ($g\,t^{-1}t^{-1}$)
1	RRII 422	33.1
2	RRII 414	86.7
3	RRII 403	55.0
4	RRII 105	41.1
5	RRIC 100	52.1
6	RRII 429	52.2
7	RRII 407	33.9
8	RRII 430	84.6
	SE.d 6.25	
	CD (p=0.05)	13.50

HEVEA BREEDING SUB-STATION, MARTHANDAM, TAMIL NADU

1. Genetic improvement of *Hevea brasiliensis* for developing ideal clones

1.1. Conventional breeding

The highlights of the results pertaining to the different experimental trials under four projects that are being pursued in the Station *viz.*, clone evaluation, hybridization and clonal selection, new generation polyclonal seed garden and participatory clone evaluation are furnished in detail hereunder for the period 2020-21.

1.1.1. Clone evaluation

a Block evaluation of selected clones of *Hevea* (2019)

The trial was laid out during October 2019 at Maruthamparai Unit of Chithar Division of ARC Ltd. wiith 11 clones *viz.*, PB 255, PB 314, IRCA 109, IRCA 111, RRII 203, RRII 414, RRII 417, RRII 422, RRII 429, RRII 430 and RRII 105.

During the period under report, vacancy enumeration was undertaken and a total of 24 causalities were replaced with the plants of the respective clones.

1.1.2. Hybridization and clonal selection

The research farm at Paraliar constitutes two breeding orchards comprising of 51 parental clones and were properly maintained. During the period under report, hand pollination works were suspended owing to the lockdown (due to COVID 19 pandemic) at the time of peak flowering. The four selections pollarded from the previous year's hybrids were maintained for the next stage of evaluation.

1.1.3. New generation polyclonal seed garden

The seed garden with nine constituent clones at New Ambady Estate was maintained well. Seedling nurseries have been raised over the earlier seasons. A couple of selections have been pollarded for further evaluation.

An effort was made to study the relationship between the size and weight of seeds on the vigour and selection of seedlings. As part of this study, the following work was done.

Around 25 kg (7000 nos.) of seedlings were collected from the polyclonal seed garden in two rounds. Seeds were sorted into three size categories *viz.*, small, medium and large. Around 150 sample seeds were taken from each category and their weights were taken. Majority of the seeds collected fell in to the medium size category.

LIBRARY AND DOCUMENTATION CENTRE

The Library and Documentation Centre attached to Rubber Research Institute of India is well maintained with a collection of 23096 books, 24755 bound volumes of periodicals, 6047 standards, 1563 reprints, 193 Theses/Dissertations and 1200 Microfiche/Microfilms. Subject bibliographies and computer based bibliographic databases of all books, research articles, standards, theses and reprints are also accessible to the users.

Library continued the information and literature support to its in-house and outside institutional users by providing reference services, current awareness services and reprographic services. During the current year, 66 books, one PhD Thesis, 2 standards and one bound journal were added to the stock. Received and registered 191 issues of journals as subscription/exchange. Complete physical verification of 23476 books and 24754 bound journals was conducted during the period.

Compiled information bulletins, *viz.*, *Documentation List* (1-2) 2020, *New Additions List of Books* 2020 and *Staff Publications list* 2020-2021. Databases were updated with the details of 66 books, 226 research articles, 39 standards, 2 thesis/ dissertation and one bound journal. Circulated 669 books, technically processed 1571 books, filed 1106 press clippings of relevant articles and provided 2841 photocopies. Library membership was issued to 34 members, reference service extended to 595 users and No Dues Certificate issued to 60 members.

As a part of sales promotion of RRII publications, Library organized the sale and distribution of 517 copies of the journal *Rubber Science* and 311 other publications including RRII Annual Report and collected Rs. 59,000 including the price of publications sold, charges for overdue on circulated books, and photocopying.

SCIENTIFIC ADVISORY COMMITTEE RECOMMENDATIONS 2020-2021

- Recommended *Calopogonium caeruleum* as an alternate cover crop in rubber plantations. This leguminous cover crop can be established under partial shade also during later immaturity period of rubber (77th SAC on 18th May 2020).
- Recommended a new water based combination fungicide with the formulation of 5% Pyraclostrobin + 55% metiram (trade name Mantram) at a concentration of 1g/L to control *Corynespora* leaf disease in rubber plantations in addition to already recommended fungicides (77th SAC on 18th May 2020).
- Accepted the findings that latex can be pre-treated with suitable agents (irrespective of whether LATZ or HA latex) prior to adding preservatives and the pre-treated latex can be stored for up to six months and good quality Ribbed Smoked Sheets can be made from it (78th SAC meeting on 16th November 2020).
- Accepted the innovation of a simple, quick and accurate method for DRC determination of field latex using 70% isopropyl alcohol (Medispirit) as the coagulant. The new method was much faster and cheaper than the conventional acid coagulation method and both methods gave identical results (78th SAC meeting on 16th November 2020).
- Accepted the recommendation that the Motorol spray oil supplied by M/s Quebec Petroleum Resources Limited, Gujarat, can be used as a carrier of oil-based copper oxychloride (COC) in rubber plantations (79th SAC on 22nd January 2021).

ANNUAL EXPENDITURE 2020-21

PLAN EXPENDITURE - STRENGTHENING RUBBER RESEARCH	Expenditure (Rs.)
A. **Research (ONE)**	
Scheme Expenditure (Res ONE)	1,20,39,480
Pay & Allowances	23,05,68,368
Wages	3,58,44,082
Sub Total	**27,84,51,930**
B. **Research (NE)**	
Scheme Expenditure (Res NE)	2,40,408
Pay & Allowances	3,54,26,770
Wages	1,23,40,161
Sub Total	**4,80,07,339**

PUBLICATIONS

RESEARCH ARTICLES

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Aswathy, C.S., Teena, A. and Roy, C.B. (2021). Mining and expression profiling of NBS-LRR genes involved in pathogen sensing and host defense in *Hevea-Phytophthora* interaction. *Indian Phytopathological Society Middle East Zone National Conference*, 27-28 January 2021, Aligarh Muslim University, Aligarh, Uttar Pradesh, India.

Deepthi, R. and Gireesh, T. (2021). Recombination breeding for growth and stem straightness using certain indigenous and exotic clones of Hevea brasiliensis. *Proceedings of NHPS International Conference on New Horizons in Plant Science*, 4-9 January 2021, University of Kerala, Thiruvananthapuram, India, pp. 176-178.

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Limiya, J., Teena, A. and Roy, C.B. (2021). Augmenting breeding for disease resistance in rubber tree - Wickham clones serve as a reliable germplasm resource to excavate diverse resistance genes. *Indian Phytopathological Society Middle East Zone National Conference*, 27-28 January 2021, Aligarh Muslim University, Aligarh, Uttar Pradesh, India.

Roy, C.B., Anu K. and Limiya, J. (2020). Virus Induced Gene Silencing: A potential tool for functional genomics studies in *Hevea brasiliensis*. *Indian Phytopathological Society South Zone National Symposium*, 1-2 December 2020, Indian Agricultural Research Institute, New Delhi, India.

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Soumyamol, and Roy, C.B. (2021). Bio-sourcing endophytes as biocontrol agents of Colletotrichum leaf disease caused by *Colletotrichum* spp. in *Hevea brasiliensis*. *Indian Phytopathological Society Middle East Zone National Conference*, 27-28 January 2021, Aligarh Muslim University, Aligarh, Uttar Pradesh, India.

Teena, A., Aswathy, C.S. and Roy, C.B. (2020). Elucidating the *role of auxin in Hevea brasiliensis in response to* Phytophthora *infection*. *Indian Phytopathological Society South Zone National Symposium*, 1-2 December 2020, Indian Agricultural Research Institute, Nilgiris, Tamil Nadu, India.

Teena, A., Aswathy, C.S. and Roy, C.B. (2021). RNA sequencing reveals orchestration of WRKY transcription factors as molecular regulators of disease response in *Hevea-Phytophthora* interaction. *Indian Phytopathological Society Middle East Zone National Conference*, 27-28 January 2021, Aligarh Muslim University, Aligarh, Uttar Pradesh, India

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AWARDS

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SCIENTIFIC AND SENIOR SUPPORTING PERSONNEL

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James Jacob, M.Sc.(Ag.), Ph.D., DIC, Ph.D.

Agronomy and Soils Division	
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P. Prasannakumari, M.Sc., Ph.D.	Senior Scientist
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C.A. Johny, B.Sc.	Farm Officer
M.S. Babu	Farm Officer
Fertilizer Advisory Group	
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Joy Varghese	Farm Officer
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E.P. Joy	Farm Officer

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Reethamma Joseph, MSc., Ph.D.	Assistant Rubber Technologist
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S. Veeraputhran, M.A., M. Phil.	Scientist
T. Siju, M.Sc. (Ag.), Ph.D.	Scientist
Joby Joseph, M.A.	Scientist

Library and Documentation Centre

N. Latha, M.Sc., M.L.I.Sc.	Documentation Officer
A.S. Ajitha, M.A., M.L.I.Sc	Senior Librarian
V.R. Sujatha, B.Sc., M.L.I.Sc.	Librarian

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P. Aneesh, M.Sc., P.G.D.C.A.	Assistant Statistician
K.A. Santhosh, M.C.A.	Assistant Systems Officer

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M.R. Anilkumar, Dip. in Inst. Tech.	Instrumentation Officer
R. Suni, M.Sc., M.Tech., MBA, B.Ed., PGDCA, PGDDI	Assistant Instrumentation Officer

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B. Jayaprakash	Assistant Secretary
N.B. Smitha	Section Officer

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M.T. Varghese, MA	Assistant Security Officer

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N.K. Suresh, MBBS	Medical Officer
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V.M. Chandran	Farm Officer
Sleeba V. Paul	Farm Officer
S.B. Sreejamol	Section Officer

Regional Research Station, Padiyoor, Kerala

C.L. Benny	Farm Officer

Regional Research Station, Guwahati, Assam

Gitali Das, M.Sc., Ph.D.	Deputy Director (RS) (upto 31.8.20)

Regional Research Station, Agartala, Tripura

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Debasis Mandal, M.Sc. Ph.D.	Senior Scientist (upto 29.2.2020)
Debabrata Ray, M.Sc. (Ag.)	Scientist
Bhaskar Datta, M.Sc. (Ag.)	Scientist
Pradip Baruah, B.Com., ICWA (I)	Assistant Director (Finance)
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Regional Research Station, Tura, Meghalaya

Ramphool Singh, M.Sc. (Ag.), Ph.D.	Senior Scientist (upt o 06.04 2021)
Merry Birth N Marak	Section Officer

Regional Experiment Station, Nagrakata, West Bengal

Jayanta Sarkar, M.Sc. (Ag.), Ph.D.	Scientist (Officer-in-charge)

Regional Research Station, Dapchari, Maharashtra

Meena Singh, M.Sc. (Ag.), Ph.D.	Senior Scientist
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Regional Soil Testing Laboratory, Adoor, Kerala (Handed over to Rubber Board managed company w.e.f. 14.08.2020)

D. Suja	Junior Scientific Officer

Regional Soil Testing Laboratory, Kanjirappally, Kerala (Handed over to Rubber Board managed company w.e.f. 14.08.2020)

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Regional Soil Testing Laboratory, Kozhikode, Kerala (Handed over to Rubber Board managed company w.e.f. 14.08.2020)

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Joseph Chacko	Junior Scientific Officer

Regional Soil Testing Laboratory, Palai, Kerala (Handed over to Rubber Board managed company w.e.f. 14.08.2020)

Sherly Jacob	Junior Scientific Officer (upto 14.8.2020)

Regional Soil Testing Laboratory, Thrissur, Kerala (Handed over to Rubber Board managed company w.e.f. 14.08.2020)

Regional Soil Testing Laboratory, Nedumangadu, Kerala (Handed over to Rubber Board managed company w.e.f. 14.08.2020)

Parvathy S. Kumar	Scientific Assistant

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REGIONAL SOIL TESTING LABORATORIES IN KERALA

(The seven Regional Soil Testing Laboratories were handed over to Rubber Board managed companies in the respective locality w.e.f. 14-08.2020)

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Research Divisions and functions

The major research divisions are Agronomy/Soils, Biotechnology, Botany, Climate Change & Ecosystem Studies, Germplasm, Latex Harvest Technology, Plant Pathology, Plant Physiology, Rubber Technology, Technical Consultancy and Economics. Studies on Clone Evaluation, Genome Analysis and DRIS Fertilisation are dealt separately.

The thrust areas of research of Agronomy/Soils Division are investigations on the nutritional requirements of rubber, irrigation, intercropping, cover crop management, weed control and the study of the rubber growing soils. Development of tissue culture and another culture system for propagation and development of transgenic plants incorporating agronomically important genes for improvement of *Hevea* are the important areas in which the Biotechnology Division is engaged. The Advanced Centre for Molecular Biology and Biotechnology (ACMBB) is a functional grouping of scientists working in the areas of Molecular Biology, Biotechnology, Genome Analysis, Molecular Physiology and Molecular Pathology. The important fields of research of the Botany Division are breeding, evaluation and selection of new clones, propagation techniques, planting methods, anatomical studies and cytogenetic investigations. The Climate Change & Ecosystems Studies Division is pursuing studies on climate change process in traditional and non-traditional rubber growing regions of India and developing information system on rubber cultivation using remote sensing (RS) platform to identify area under rubber cultivation and suitable area where rubber plantations can be extended. The Germplasm Division is concentrating on the introduction, conservation and evaluation of *Hevea* germplasm. The Plant Pathology Division is engaged in investigations on the diseases and pests of rubber and associated cover crops and their control. The Plant Physiology Division conducts studies on both fundamental and applied aspects of *Hevea* tree physiology. The Latex Harvest Technology Division is concentrating on all applied aspects of crop harvesting in rubber. The Rubber Technology Division concentrates on improvement in primary processing of rubber, its chemical modification, rubber product manufacture and quality control of processed rubber. The Technical Consultancy Division provides consultancy services for the promotion of the rubber industry. The Rubber Technology Division and Technical consultancy Division together forms the Advanced Centre for Rubber Technology (ACRT). The Economics Division undertakes studies on economic aspects related to rubber plantations.

The research supporting sections includes Library and Documentation, Instrumentation, Statistics, Computer and Maintenance Wing. There is also a small experimental farm of 33 ha. at the headquarters of RRII.

Central Experiment Station

The 255 ha. Central Experiment Station at Chethackal (Ranni), 50 km away from Kottayam, was started in 1966. Field trials laid out by the research divisions cover almost the entire area.

Regional Research Stations

RRII has established a North-Eastern Research Complex with headquarters at Agartala having regional research stations at Agartala in Tripura, Guwahati in Assam and Tura in Meghalaya. The RRII has also set up regional research establishments at Dapchari (Maharashtra), Dhenkanal (Orissa), Nagrakata (West Bengal), Thadikarankonam (Tamil Nadu), Kadaba (Karnataka) and Padiyoor (Kerala).

Regional soil testing laboratories have been established at Kozhikode, Thrissur, Muvattupuzha, Pala, Kanjirappally, Adoor and Nedumangad. Mobile units for soil and leaf analysis are available at Kozhikode laboratory, apart from that at the headquarters.

National/International collaboration

RRII is a member of the International Rubber Research and Development Board (IRRDB), an association of national organizations devoted to research and development on natural rubber. Rubber Board is a member of the Association of Natural Rubber Producing Countries (ANRPC) and International Rubber Study Group (IRSG).

The RRII has research/academic linkages with the Banaras Hindu University (Varanasi), Kerala Agricultural University (Thrissur), Kerala University (Thiruvananthapuram), Mahatma Gandhi University (Kottayam), Cochin University of Science and Technology (Kochi), Indian Agricultural Research Institute (New Delhi), Indian Institute of Sciences (Bangalore), Indian Institute of Technology (Kharagpur), National Chemical Laboratory (Pune), Sree Chitra Tirunal Institute of Medical Sciences and Technology (Thiruvananthapuram), Tamil Nadu Agricultural University (Coimbatore), University of Agricultural Sciences (Bangalore) and University of Goa (Goa).

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