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**CROP HUSBANDRY (5 jdl)**

P. Dissanayake, D.L. George, M.L. Gupta, Direct seeding as an alternative to transplanting for guayule in southeast Queensland,

***Industrial Crops and Products***, Volume 27, Issue 3, May 2008, Pages 393-399, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2007.11.001.

(<http://www.sciencedirect.com/science/article/pii/S0926669007001458>)

**Abstract:**

Guayule (*Parthenium argentatum* Gray) is a source of high quality rubber and low-allergenic latex. Commercial potential of guayule to produce high value latex products has increased due to the increased incidence of deadly diseases in humans. The objective of this study was to investigate the potential of direct seeding in southeast Queensland as an alternative to establishment by the current high cost transplanting method. Experiments were conducted for 2 years at Gatton, Queensland using physical and chemical seed treatments. NaOCl plus GA3, osmo-priming with polyethylene glycol, seed pelleting and seed tape planting were tested. Planting depth had a significant influence on seedling emergence. Emergence at 18 mm depth (1.5 seedlings/m) was significantly reduced compared with 10 mm depth (2.9 seedlings/m). Osmo-priming was effective in increasing germination from 36 to 47%. It also improved seedling emergence and vigour. Osmo-priming significantly increased establishment (7.5 seedlings/m) compared with untreated seed (3.3 seedlings/m). NaOCl plus GA3 did not have a significant influence on germination (38%) or seedling emergence (5.0 seedlings/m) but increased survival at 42 days after planting. Establishment as a percentage of emerged seedlings was high for both osmo-primed and NaOCl plus GA3 treatments with 89 and 88% respectively, whereas untreated seed had only 70% survival. Osmo-priming also increased vigour as indicated by increased root length (101 mm), shoot height (123 mm), and seedling dry matter (379 mg/seedling) compared with the control (83, 107 mm and 206 mg/seedling, respectively).

**Keywords:** Guayule; *Parthenium argentatum*; Establishment; Direct seeding; Osmo-priming; Pelleting; GA3

G.M. Bedane, M.L. Gupta, D.L. George, Effect of plant population on seed yield, mass and size of guayule,

***Industrial Crops and Products***, Volume 29, Issue 1, January 2009, Pages 139-144, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2008.04.020.

(<http://www.sciencedirect.com/science/article/pii/S0926669008000824>)

**Abstract:**

Guayule (*Parthenium argentatum* Gray) is a source of high quality low-allergenic natural rubber. It is a relatively new crop in Australia and optimum plant density for seed

production has not been established. The objective of the current experiment was to examine whether seed yield, mass and size are affected by plant population. The effect of plant population on seed yield, mass and size was investigated by planting guayule (AZ-2) at 4444, 8300, 12,500 and 25,000 plants/ha. Data were collected at 16 and 28 months after planting. Seed was harvested manually multiple times over 4 weeks each year following the main flowering period in spring. Harvested seed was threshed and clean seed yield was compared among different plant populations. Seed quality attributes were also compared in terms of 1000-seed mass and seed size. Lowest plant population of 4444 plants/ha provided the highest yield at 28 months but was the lowest yielding at 16 months because the plants had not yet reached full size to compensate for the wider spacing. However, at both ages this treatment produced heavier and larger seeds. The difference in yield or seed mass and size between plant populations ranging from 8300 to 25,000 plants/ha was not significant. Overall results of the study demonstrated that seed yield and seed size, which is important in direct seeding could be affected by plant population.

**Keywords: Guayule; Plant population; Yield; Seed size classes; 1000-Seed mass**

G.M. Bedane, M.L. Gupta, D.L. George, Optimum harvest maturity for guayule seed, *Industrial Crops and Products*, Volume 24, Issue 1, July 2006, Pages 26-33, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2005.10.005.

(<http://www.sciencedirect.com/science/article/pii/S0926669005001184>)

**Abstract:**

Guayule (*Parthenium argentatum* Gray) is a rubber-producing shrub native to the semi-arid region of north central Mexico and southwestern Texas. Timely harvest is critical to achieve maximum seed viability, vigour, and yield. The objective of this study was to investigate possible indicators of optimum seed maturity in guayule. The optimum harvest maturity time for guayule was studied by comparing quality parameters at different times after flowering. Heat units expressed as growing degree-days after flowering were calculated and related to seed development stages and quality. Seed quality at different stages of development was assessed by germination, capitulum dry mass, 1000 seed mass, and percentage of filled seeds. The maximum seed quality was recorded at 329 growing degree-days (GDD). This was 28 days from time of flowering. At this date, the moisture content of the capitulum was 48% on a wet basis and the colour was comparable to cinnamon (Code 165C) on the Royal Horticultural Society (R.H.S.) standard colour chart. Of all the parameters GDD, 1000 seed mass, and percentage of filled seeds provided a more rapid and reliable measure of optimum seed maturity. Colour identification can be used as an additional indicator.

**Keywords: Guayule; Parthenium argentatum; Seed quality; Timely harvest; Degree-days; Seedhead colour; Shattering**

Panida Kongsawadworakul, Unchera Viboonjun, Phayao Romruensukharom, Pisamai Chantuma, Somjintana Ruderman, Herve Chrestin, The leaf, inner bark and latex cyanide potential of *Hevea brasiliensis*: Evidence for involvement of cyanogenic glucosides in rubber yield,

***Phytochemistry***, Volume 70, Issue 6, April 2009, Pages 730-739, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2009.03.020.

(<http://www.sciencedirect.com/science/article/pii/S0031942209001344>)

**Abstract:**

The latex of *Hevea brasiliensis*, expelled upon bark tapping, is the cytoplasm of anastomosed latex cells in the inner bark of the rubber tree. Latex regeneration between two tappings is one of the major limiting factors of rubber yield. *Hevea* species contain high amounts of cyanogenic glucosides from which cyanide is released when the plant is damaged providing an efficient defense mechanism against herbivores. In *H. brasiliensis*, the cyanogenic glucosides mainly consist of the monoglucoside linamarin (synthesized in the leaves), and its diglucoside transport-form, linustatin. Variations in leaf cyanide potential (CNp) were studied using various parameters. Results showed that the younger the leaf, the higher the CNp. Leaf CNp greatly decreased when leaves were directly exposed to sunlight. These results allowed us to determine the best leaf sampling conditions for the comparison of leaf CNp. Under these conditions, leaf CNp was found to vary from less than 25 mM to more than 60 mM. The rubber clones containing the highest leaf CNp were those with the highest yield potential. In mature virgin trees, the CNp of the trunk inner bark was shown to be proportional to leaf CNp and to decrease on tapping. However, the latex itself exhibited very low (if any) CNp, while harboring all the enzymes ([beta]-d-diglucosidase, linamarase and [beta]-cyanoalanine synthase) necessary to metabolize cyanogenic glucosides to generate non-cyanogenic compounds, such as asparagine. This suggests that in the rubber tree bark, cyanogenic glucosides may be a source of buffering nitrogen and glucose, thereby contributing to latex regeneration/production.

**Keywords:** *Hevea brasiliensis*; Euphorbiaceae; Rubber tree; Latex; Leaf and bark cyanide potential; Cyanide metabolism; Linamarin; Rubber yield

Calvin H. Pearson, Katrina Cornish, Colleen M. McMahan, Donna J. Rath, Jenny L. Brichta, Jennifer E. Van Fleet, Agronomic and natural rubber characteristics of sunflower as a rubber-producing plant,

***Industrial Crops and Products***, Volume 31, Issue 3, May 2010, Pages 481-491, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2010.01.010.

(<http://www.sciencedirect.com/science/article/pii/S0926669010000130>)

**Abstract:**

Sunflower (*Helianthus annuus* L., Asteraceae) is a genus native to North America and is a potential natural rubber (NR) producing crop. The objectives of the study were to: (1) evaluate commercial sunflower cultivars to determine biomass production and how they partition biomass into leaves, stems, and head, (2) determine how removing the head affects biomass partitioning, (3) determine latex concentration and yield in commercial sunflower cultivars and a diversity of genetic sunflower material, and (4) characterize

several quality factors pertaining to the latex produced by commercial sunflower cultivars and a diversity of genetic sunflower material. Field performance tests were conducted at the Western Colorado Research Center at Fruita, Colorado for three growing seasons (2001, 2002, and 2003). Latex was found almost entirely in the leaves of young and mature sunflowers. No latex was found in mature stems or in the pappus of the flowers. On average, sunflower partitioned biomass into 18% leaves, 38% stems, and 44% heads. With the head removed, sunflower partitioned biomass into 33% leaves and 67% stems. Sunflower cultivars exhibited considerable genetic variation for biomass partitioning and also for NR yield and quality. Sunflower synthesized NR with 95-97% being low molecular weights ranging from 66,000 to 74,000 g/mol and a small, remarkable percentage (~5%) of NR being higher molecular weight (~600,000 g/mol). The potential for increasing latex production in sunflower appears possible, given that current NR levels are low and reasonable advances in NR production in sunflower plants through plant breeding and genetic engineering might be achieved. Based on the genetics of the sunflower cultivars included in our study, it appears that some cultivars would be more responsive to plant breeding for increased leaf mass than others. The development of sunflower cultivars suitable for commercial production of NR will require significant improvements in the quantity and quality of NR produced in the plant.

**Keywords: Natural rubber; Sunflower; Natural rubber plants; Rubber quality; Natural rubber extraction**

## **PLANT PROPAGATION (4 jdl)**

T.A. Coffelt, F.S. Nakayama, D.T. Ray, K. Cornish, C.M. McMahan, C.F. Williams, Plant population, planting date, and germplasm effects on guayule latex, rubber, and resin yields,

***Industrial Crops and Products***, Volume 29, Issue 1, January 2009, Pages 255-260, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2008.05.010.

(<http://www.sciencedirect.com/science/article/pii/S0926669008001209>)

### **Abstract:**

Guayule (*Parthenium argentatum* Gray) is a perennial shrub native to the Chihuahuan Desert. While guayule traditionally has been cultivated for rubber, more recently it is being cultivated for its hypoallergenic latex. Other uses including termite resistant wood products and an energy source have also been identified. However, the effects of various agronomic practices, such as planting and harvesting dates, plant spacing, cutting height and frequency, irrigation frequency, and herbicide application, on latex concentration and yield of newly developed germplasm have not been reported. The objectives of this study were to determine the yield and concentration of latex, rubber, and resin of four guayule lines planted at two populations and two planting dates. Four guayule lines (AZ-1, AZ-3, AZ-5, and 11591) were transplanted at two dates (28 November 2000 and 7 June 2001) and two plant populations (27,000 and 54,000 plants ha<sup>-1</sup>). Treatments were replicated four times. Each treatment plot was subdivided into six subplots for harvesting at 6-month intervals beginning 1 year after transplanting. Results showed that transplanting date did not affect plant size or latex concentration or

yield consistently. Instead, it appeared that the time of harvest (fall vs. spring) was more important. The sixth (last harvest) in the fall planting date and the fifth harvest date in the spring planting date were the optimum for plant biomass and latex, rubber, and resin concentrations and yields. The lines AZ-1 and AZ-3 were larger, whereas AZ-5 had higher latex and rubber concentrations than the control, 11591. The greater plant population (54,000 plants ha<sup>-1</sup>) had higher biomass, rubber, and resin yields than the lower population (27,000 plants ha<sup>-1</sup>) at the early harvest dates, but not at the later harvest dates (5 and 6). More studies must be conducted to determine the optimum plant population and transplanting date for other newly developed guayule germplasm lines.

**Keywords: Plant population; Planting date; Germplasm; Latex; Rubber; Resin; Yield; Parthenium argentatum; Guayule; Agronomic practices**

T.A. Coffelt, D.T. Ray, Cutting height effects on guayule latex, rubber, and resin yields, *Industrial Crops and Products*, Volume 32, Issue 3, November 2010, Pages 264-268, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2010.04.018.

(<http://www.sciencedirect.com/science/article/pii/S0926669010001093>)

**Abstract:**

Guayule (*Parthenium argentatum* Gray) is a perennial shrub native to the Chihuahuan Desert. While guayule traditionally has been cultivated for rubber, more recently it is being cultivated for its hypoallergenic latex. Other uses including termite resistant wood products and as an energy source have also been identified. Major advances have been made since 1970 in the development of improved guayule germplasm. However, the effects of harvesting practices such as cutting height and frequency on latex concentration and yield of newly developed germplasm have not been reported. These results are needed in order to develop production schemes for successful commercialization of guayule. The objectives of this study were to determine the yield and concentration of latex, rubber, and resin of five guayule lines harvested at two cutting heights over 3 years and five harvesting schedules. Harvesting at 100% of plant height gave higher yields than harvesting at 50% of plant height independent of the harvest schedule. Harvesting at 100% after 4 years of growth gave the highest yields, but more research is needed to determine if harvesting on a 2-year schedule may be better for harvesting equipment and extraction equipment. AZ-2 and AZ101 were the largest plants, while N9-3 and 11591 were smaller with AZ-1 intermediate. As indicated in previous studies, the environment plays a large role in determining biomass, latex, rubber, and resin yields in guayule. In the future, optimum harvesting schemes at 100% of plant height may need to be developed for each line and environment.

**Keywords: Harvest date; Germplasm; Latex; Rubber; Resin; Biomass; Parthenium argentatum; Guayule; Agronomic practices**

Michael E. Salvucci, Terry A. Coffelt, Katrina Cornish, Improved methods for extraction and quantification of resin and rubber from guayule, ***Industrial Crops and Products***, Volume 30, Issue 1, July 2009, Pages 9-16, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2008.12.006. (<http://www.sciencedirect.com/science/article/pii/S0926669008002367>)

**Abstract:**

Guayule, a shrub native to the Chihuahuan desert, is a natural source of high quality, hypoallergenic rubber. Unlike rubber trees that produce rubber in laticifers, the rubber in guayule is produced in parenchyma cells of the bark tissue of stems and roots. Consequently, guayule tissue must be mechanically broken before the rubber can be extracted and analyzed. Since rubber extraction and analysis is time-consuming, progress towards increasing the rubber content of guayule through breeding or better cultivation practices has been limited by the slow rate of sample processing. To address the need for faster and more efficient sample throughput, conditions were optimized for automated extraction of dried guayule tissue using accelerated solvent extraction (ASE) and rapid methods were developed to replace gravimetric determination of resin and rubber content. For resin analysis, ultraviolet absorbance was used to determine resin concentration after ASE of the tissue with acetone or acetonitrile. For rubber analysis, evaporative light scattering (ELS) was used to determine the amount of rubber recovered after ASE of the tissue with cyclohexane. Extraction of guayule tissue with high latex rubber content verified that the amounts of resin and rubber determined by these methods were similar to the amounts determined gravimetrically. Since these methods automate extraction and increase the speed of resin and rubber quantification, they could be used in combination with ASE to increase the throughput and efficiency of guayule evaluation in germplasm enhancement and agronomic improvement programs.

**Keywords:** Accelerated solvent extraction; Evaporative light scattering; Guayule; *Parthenium argentatum*; Resin; Rubber

Wilaiwan Chotigeat, Sarapee Duangchu, Rapepun Wititsuwannakun, Amornrat Phongdara, Cloning and characterization of pectate lyase from *Hevea brasiliensis*, ***Plant Physiology and Biochemistry***, Volume 47, Issue 4, April 2009, Pages 243-247, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2008.12.008. (<http://www.sciencedirect.com/science/article/pii/S0981942808002374>)

**Abstract:**

Latex from the commercial *Hevea brasiliensis* contains 30-50% (w/w) of natural rubber (cis-1,4-polyisoprene), the raw material for the many products of the rubber industry. We have constructed a cDNA library from the latex of *H. brasiliensis* to investigate the expressed genes and molecular events in the latex. We have isolated two cDNAs from this library, Hb-PEL-1 and Hb-PEL-2 that could encode for pectate lyase enzymes (EC4.2.2.2). From their sequence analysis Hb-PEL-1 and Hb-PEL-2 encode for proteins of 393 and 323 amino acids, respectively. Comparison of these deduced amino acid sequences with other pectate lyase enzymes showed they contained the conserved NADPH, Ca<sup>2+</sup> and substrate binding sites and had a 74% identity to *Arabidopsis thaliana* pectate lyase. Only the Hb-PEL-1 recombinant protein expressed from *Escherichia coli* had enzymic activity which was Ca<sup>2+</sup> dependent. Interestingly, Hb-PEL-1 contained an extra internal peptide between amino acid residue 38-108 when

compared to Hb-PEL-2 and this peptide was also present in other pectate lyase enzymes. The transcript of pectate lyase (Hb-PEL) in the latex of rubber tree at various times after the first tapping was quantified by real-time PCR using 18s genes as internal standard. Most transcripts were detected on the first day after tapping and then decreased with time. This indicates that the pectate lyase may be involved in either the release of latex by breaking down the laticifer wall or in the development of laticifers.

**Keywords: cDNA library; Hevea brasiliensis; Latex; Pectate lyase; Rubber tree**

## **SOIL CULTIVATION (1 jdl)**

P. Servadio, Applications of empirical methods in central Italy for predicting field wheeled and tracked vehicle performance,

***Soil and Tillage Research***, Volume 110, Issue 2, November 2010, Pages 236-242, ISSN 0167-1987, DOI: 10.1016/j.still.2010.08.009.

(<http://www.sciencedirect.com/science/article/pii/S0167198710001492>)

### **Abstract:**

Due to the agricultural field traffic and to the soil tillage implements, soil compaction has been recognised as a severe problem in mechanized agriculture and has an influence on many soil properties and processes. In this paper, empirical methods originally developed by the different Authors at Waterways Experiment Station (WES) for predicting the performance of off-road vehicles were applied on agricultural soil. The models considered and based on soil-vehicles interaction were: clay-tire rubber track numeric (Nc,r), mobility index (MI), vehicle cone index (VCI), mean maximum pressure (MMP) and they included besides soil strength, the load carried by the tire or track, some technical characteristics of the tire or track of the vehicle, as well as the number of passes on the same track. These models have been validated with the tests results of a number of selected agricultural and forest vehicles over a range of soil in central Italy: Vertic Cambisol, Haplic Calcaric Cambisol and Eutric Cambisol. Significant correlations among the above indexes and among Nc,r and two tire-track performance parameters: traction coefficient and traction efficiency, have been found. In addition a correlation between the measured cone index values during field tests, and the predicted cone index values have been developed. Through the field data collected and the elaboration and validation of the indexes it was possible to frame in a coherent way, the performance of agricultural machinery of different mass and power, running gear system (wheels, tires with low aspect ratio, metal and rubber tracks, self-propelled for the harvest, transport and distribution of agricultural products), even though particular interpretative shrewdness was necessary in the application of such formulas in the cases of innovative machinery as with the rubber track system. The vehicles that have obtained the higher values of Nc,r (>20) have obtained the lowest values of both the MMP (<156 kPa) and VCI (VCI1 < 110 kPa; VCI4 < 269 kPa). These vehicles have also obtained the higher values of traction coefficient (0.7-0.9) and of traction efficiency (0.74-0.8). Agricultural field traffic is an important aspect of soil management and such indexes, based on parameters that determine the impact of the agricultural

mechanization on soil qualities, can usefully be considered in the management of the agricultural farm to reduce soil compaction due to both the traffic of machinery and to tillage implements particularly when considering the aspect of altered land use.

**Keywords: Wheeled tractor; Tracked tractor; Soil cone index; Predicting wheeled vehicle performance; Predicting tracked vehicle performance; Central Italy**

## **CROPPING PATTERNS AND SYSTEMS (1 jdl)**

S.M.M. Iqbal, C.R. Ireland, V.H.L. Rodrigo, A logistic analysis of the factors determining the decision of smallholder farmers to intercrop: A case study involving rubber-tea intercropping in Sri Lanka,

***Agricultural Systems***, Volume 87, Issue 3, March 2006, Pages 296-312, ISSN 0308-521X, DOI: 10.1016/j.agsy.2005.02.002.

(<http://www.sciencedirect.com/science/article/pii/S0308521X05000429>)

### **Abstract:**

The factors influencing the decision of smallholder farmers to adopt new farming technologies were studied with reference to rubber-tea intercropping in Sri Lanka. Rubber-tea intercropping has been recommended previously to rubber farmers as a means to improve productivity and income during the early pre-tapping phase of rubber growth. Although crop trials have shown that the two crops are agronomically compatible and potentially produce a combined economic yield superior to the yield of a sole crop grown on the same area of land, there is little evidence of widespread adoption of this practice among smallholder farmers in Sri Lanka. The aim of the study was to determine the major factors that influence the decision to undertake rubber-tea intercropping and to construct a predictive model that describes the likelihood of adoption of intercropping by traditional smallholder rubber growers. A rapid rural appraisal (RRA) was undertaken based on semi-structured interviews of 90 smallholder farmers in the main rubber growing low wet zone of Sri Lanka. Among a number of factors shown to significantly influence the decision to intercrop tea with rubber, three were shown to operate independently, namely level of income, source of income (i.e. solely from own farm or from farm plus additional off-farm enterprises), and availability of land considered suitable for tea cultivation. A statistical model developed through correlation and logistic analysis, which predicts the likelihood of a smallholder adopting intercropping based on these factors, is presented and discussed. The most likely combination of circumstances (82% probability) under which rubber-tea intercropping is practiced is shown to be where the farmer's income is greater than Rs. 10,000 per month, where the farmer's income is based solely on own farm enterprises, and where more than 80% of the farmer's land area was judged to be suitable for tea cultivation. Conversely, 30% of smallholder farmers that chose not to intercrop did possess land suitable for tea cultivation. Qualitative responses to the RRA indicated that limitation of technical knowledge was the main problem subsequently faced by rubber farmers who had adopted rubber-tea intercropping. Results indicate that there is need for both



income support through farm subsidies and further agricultural extension services, if rubber-tea intercropping is to be adopted more widely in Sri Lanka. The wider usefulness of the developed logistic model in determining the likelihood of adoption of intercropping by smallholder farmers is discussed.

**Keywords:** Rubber; Tea; Intercropping; Logistic model; Rapid rural appraisal

## **PLANT BREEDING AND GENETICS (5 jdl)**

V.H. Teetor, D.T. Ray, W.W. Schloman Jr., Evaluating chemical indices of guayule rubber content: Guayulins A and B, *Industrial Crops and Products*, Volume 29, Issues 2-3, March 2009, Pages 590-598, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2008.11.005. (<http://www.sciencedirect.com/science/article/pii/S0926669008002094>)

### **Abstract:**

Guayule (*Parthenium argentatum* Gray) is now a commercial crop for the production of high-quality, hypoallergenic natural rubber latex. Because guayule is relatively resistant to both insect and disease pests, its cultivation requires little chemical input. It has been postulated that guayule's chemical defense system is based on terpene derivatives such as guayulins A and B. The goals of this research were to: (1) describe the distribution of guayulins A and B throughout the plant, (2) determine whether guayulins can be used as a predictor of rubber content/yield (is guayulin content related to rubber content), and (3) determine whether the guayulin content/distribution has been changed through plant breeding. Two plants each of three different lines (11591, AZ-1, and AZ-3) and two ages (1 and 2 years old) were harvested at the soil line in the fall of 2002 and spring of 2003. Plants were separated into eight parts: brown leaves, green leaves, stem tips, stems less than 5 mm in diameter, stems between 5 and 10 mm, stems greater than 10 mm, green stems, and flower parts. Samples were analyzed for guayulins A and B and rubber content. Guayulins A and B and rubber were found in all plant parts, but were most prevalent in stems larger than 10 mm in diameter, and were significantly correlated with each other in these stems. Guayulin A was found in greater concentrations than guayulin B. There were no significant differences between plant ages for rubber, or guayulin content at the time of harvest, or guayulins for harvest season. Rubber percent was higher in spring, which agrees with numerous other investigations. Guayulin A in the stems is correlated with the total rubber in the plant, suggesting it as a potential selection tool. However, rubber in the same plant parts has a higher correlation with total plant rubber and remains a better and easier selection criterion than guayulin A. The extent to which guayulin contents differed between older, relatively unimproved lines and newer improved lines was evaluated. Variety 11591 had a higher concentration of guayulin A than guayulin B, so that the ratio of A to B was much higher than in the two improved lines (AZ-1 and AZ-3). It appears that the ratio of guayulin A to guayulin B has been changed in the two newer selections, but it is unclear whether this will affect pest resistance.

**Keywords:** Guayule; Latex; *Parthenium argentatum*; cis-1,4-Polyisoprene; Rubber; Guayulin A; Guayulin B

Calvin H. Pearson, Katrina Cornish, Colleen M. McMahan, Donna J. Rath, Jenny L. Brichta, Jennifer E. Van Fleet, Agronomic and natural rubber characteristics of sunflower as a rubber-producing plant,

***Industrial Crops and Products***, Volume 31, Issue 3, May 2010, Pages 481-491, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2010.01.010.

(<http://www.sciencedirect.com/science/article/pii/S0926669010000130>)

**Abstract:**

Sunflower (*Helianthus annuus* L., Asteraceae) is a genus native to North America and is a potential natural rubber (NR) producing crop. The objectives of the study were to: (1) evaluate commercial sunflower cultivars to determine biomass production and how they partition biomass into leaves, stems, and head, (2) determine how removing the head affects biomass partitioning, (3) determine latex concentration and yield in commercial sunflower cultivars and a diversity of genetic sunflower material, and (4) characterize several quality factors pertaining to the latex produced by commercial sunflower cultivars and a diversity of genetic sunflower material. Field performance tests were conducted at the Western Colorado Research Center at Fruita, Colorado for three growing seasons (2001, 2002, and 2003). Latex was found almost entirely in the leaves of young and mature sunflowers. No latex was found in mature stems or in the pappus of the flowers. On average, sunflower partitioned biomass into 18% leaves, 38% stems, and 44% heads. With the head removed, sunflower partitioned biomass into 33% leaves and 67% stems. Sunflower cultivars exhibited considerable genetic variation for biomass partitioning and also for NR yield and quality. Sunflower synthesized NR with 95-97% being low molecular weights ranging from 66,000 to 74,000 g/mol and a small, remarkable percentage (~5%) of NR being higher molecular weight (~600,000 g/mol). The potential for increasing latex production in sunflower appears possible, given that current NR levels are low and reasonable advances in NR production in sunflower plants through plant breeding and genetic engineering might be achieved. Based on the genetics of the sunflower cultivars included in our study, it appears that some cultivars would be more responsive to plant breeding for increased leaf mass than others. The development of sunflower cultivars suitable for commercial production of NR will require significant improvements in the quantity and quality of NR produced in the plant.

**Keywords: Natural rubber; Sunflower; Natural rubber plants; Rubber quality; Natural rubber extraction**

M.E. Veatch-Blohm, D.T. Ray, A. Gehrels, Night temperature, rubber production, and carbon exchange in guayule,

***Industrial Crops and Products***, Volume 25, Issue 1, January 2007, Pages 34-43, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2006.06.019.

(<http://www.sciencedirect.com/science/article/pii/S092666900600094X>)

**Abstract:**

Guayule (*Parthenium argentatum* Gray), a native of the Chihuahuan desert, produces the majority of its rubber during the winter months. Increased rubber production is thought to be induced by cold night temperatures, but the factors involved in rubber induction are not completely understood. The purpose of this study was to answer three questions about rubber production in the winter: (1) how do immature plants (<180

days) respond to cold night temperatures?; (2) how is rubber production in mature plants ([approximate]1 year) related to night temperature?; and (3) what is the relationship between carbon exchange and rubber production under cold night temperatures? Plants were grown in differentially heated enclosures over three consecutive winters. The warm-night plants were exposed to simulated summer night temperatures and the cold-night plants to ambient winter night temperatures. Plant responses to different night temperatures were monitored by measuring growth, carbon exchange, fresh and dry weight, and resin and rubber production. Immature plants exposed to cold nights had higher rubber concentration than the warm-night plants only in the first year, which had the lowest night temperatures. For the mature plants, dry weight was not significantly different between treatments, but rubber concentration and yield were significantly greater in the cold-night than the warm-night plants. Plants in both treatments had similar carbon exchange rates. Therefore, the similarity in dry weight between treatments was most likely due to increased growth in the warm-night plants and increased rubber deposition in the cold-night plants. Rubber concentration was significantly related to night temperature in both mature and immature plants and appears to be stimulated most by temperatures below 10 [degree sign]C.

**Keywords: Guayule; Night temperature; Rubber induction; Carbon exchange**

P. Dissanayake, D.L.George, M.L. Gupta, Improved guayule lines outperform old lines in south-east Queensland,

*Industrial Crops and Products*, Volume 25, Issue 2, February 2007, Pages 178-189, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2006.09.002.

(<http://www.sciencedirect.com/science/article/pii/S0926669006001245>)

**Abstract:**

Guayule (*Parthenium argentatum* Gray) is a source of high quality rubber and low-allergenic latex as well as resin for use as a wood preservative. Demand for high value latex products has increased with the advent of deadly diseases such as AIDS. The objective of this study was to evaluate the performance of six improved guayule lines (AZ-1 to AZ-6) in south-east Queensland: released jointly by the Agricultural Research Service (ARS) of the United States Department of Agriculture (USDA) and The University of Arizona. Trials were conducted at two sites, Chinchilla and Gatton. Overall performance of improved lines for plant growth and yield of dry matter, rubber and resin was better at both Gatton and Chinchilla than the standard check lines (N 565 and 11591). AZ-1 and AZ-2 maintained the best combinations of desirable traits, including plant uniformity, early vigorous growth, increased dry matter, and increased rubber and resin yields. Of these two, AZ-2 had more uniform plant growth and has commercial potential for Queensland production areas. In the summer harvest at Gatton, 32-month-old AZ-1 and AZ-2 produced rubber yields of 789 kg/ha and 771 kg/ha, respectively, while controls, N 565 and 11591 produced 675 kg/ha and 618 kg/ha, respectively. At Chinchilla, at 33 months, spring harvested AZ-1 and AZ-2 produced rubber yields of 717 kg/ha and 787 kg/ha; these yields were significantly higher than N 565 and 11591 which produced 385 kg/ha and 380 kg/ha, respectively. Thus, rubber yields of AZ-1 and AZ-2 were consistently high across sites. AZ-1 and AZ-2 produced resin yields of 1158 kg/ha and 1115 kg/ha at Gatton and 1318 kg/ha and 1476 kg/ha at Chinchilla. This compared

with a mean of 612 kg/ha and 352 kg/ha for the standard check lines at Gatton and Chinchilla. Thus resin yields of AZ-1 and AZ-2 were consistently high across sites. Rubber content appeared to be influenced by time of harvest although this effect is compounded with plant age. At Gatton, in spring, 17-month-old plants produced a mean rubber content of 7.7% (all lines), while, in summer, when the plants were 32-month-olds, rubber content dropped to 6.4%. At Chinchilla, 33-month-old plants harvested in spring produced a mean rubber content of 7.4%, similar to the spring value at Gatton. By contrast, resin content appeared to be little affected by season.

**Keywords: Guayule; Parthenium argentatum; Improved lines; Evaluation; Rubber; Resin**

T.A. Coffelt, F.S. Nakayama, D.T. Ray, K. Cornish, C.M. McMahan, C.F. Williams, Plant population, planting date, and germplasm effects on guayule latex, rubber, and resin yields,

*Industrial Crops and Products*, Volume 29, Issue 1, January 2009, Pages 255-260, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2008.05.010.

(<http://www.sciencedirect.com/science/article/pii/S0926669008001209>)

**Abstract:**

Guayule (*Parthenium argentatum* Gray) is a perennial shrub native to the Chihuahuan Desert. While guayule traditionally has been cultivated for rubber, more recently it is being cultivated for its hypoallergenic latex. Other uses including termite resistant wood products and an energy source have also been identified. However, the effects of various agronomic practices, such as planting and harvesting dates, plant spacing, cutting height and frequency, irrigation frequency, and herbicide application, on latex concentration and yield of newly developed germplasm have not been reported. The objectives of this study were to determine the yield and concentration of latex, rubber, and resin of four guayule lines planted at two populations and two planting dates. Four guayule lines (AZ-1, AZ-3, AZ-5, and 11591) were transplanted at two dates (28 November 2000 and 7 June 2001) and two plant populations (27,000 and 54,000 plants ha<sup>-1</sup>). Treatments were replicated four times. Each treatment plot was subdivided into six subplots for harvesting at 6-month intervals beginning 1 year after transplanting. Results showed that transplanting date did not affect plant size or latex concentration or yield consistently. Instead, it appeared that the time of harvest (fall vs. spring) was more important. The sixth (last harvest) in the fall planting date and the fifth harvest date in the spring planting date were the optimum for plant biomass and latex, rubber, and resin concentrations and yields. The lines AZ-1 and AZ-3 were larger, whereas AZ-5 had higher latex and rubber concentrations than the control, 11591. The greater plant population (54,000 plants ha<sup>-1</sup>) had higher biomass, rubber, and resin yields than the lower population (27,000 plants ha<sup>-1</sup>) at the early harvest dates, but not at the later harvest dates (5 and 6). More studies must be conducted to determine the optimum plant population and transplanting date for other newly developed guayule germplasm lines.

**Keywords: Plant population; Planting date; Germplasm; Latex; Rubber; Resin; Yield; Parthenium argentatum; Guayule; Agronomic practices**

## PLANT PHYSIOLOGY AND BIOCHEMISTRY (34 jdl)

Wannapa Sritanyarat, Gregory Pearce, William F. Siems, Clarence A. Ryan, Rapepun Wititsuwannakul, Dhirayos Wititsuwannakul, Isolation and characterization of iso inhibitors of the potato protease inhibitor I family from the latex of the rubber trees, *Hevea brasiliensis*,

**Phytochemistry**, Volume 67, Issue 15, Rod Croteau Special Issue, Part 1, August 2006, Pages 1644-1650, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2005.12.016.

(<http://www.sciencedirect.com/science/article/pii/S0031942205006795>)

### **Abstract:**

Three iso inhibitors have been isolated to homogeneity from the C-serum of the latex of the rubber tree, *Hevea brasiliensis* clone RRIM 600, and named HPI-1, HPI-2a and HPI-2b. The three inhibitors share the same amino acid sequence (69 residues) but the masses of the three forms were determined to be 14,893 +/- 10, 7757 +/- 5, and 7565 +/- 5, respectively, indicating that post-translational modifications of the protein have occurred during latex collection. One adduct could be removed by reducing agents, and was determined to be glutathione, while the other adduct could not be removed by reducing agents and has not been identified. The N-termini of the inhibitor proteins were blocked by an acetylated Ala, but the complete amino acid sequence analysis of the deblocked inhibitors by Edman degradation of fragments from endopeptidase C digestion and mass spectrometry confirmed that the three iso inhibitors were derived from a single protein. The amino acid sequence of the protein differed at two positions from the sequence deduced from a cDNA reported in GenBank. The gene coding for the inhibitor is wound-inducible and is a member of the potato inhibitor I family of protease inhibitors. The inhibitor strongly inhibited subtilisin A, weakly inhibited trypsin, and did not inhibit chymotrypsin. The amino acid residues at the reactive site P1 and were determined to be Gln45 and Asp46, respectively, residues rarely reported at the reactive site in potato inhibitor I family members. Comparison of amino acid sequences revealed that the HPI iso inhibitors shared from 33% to 55% identity (50-74% similarity) to inhibitors of the potato inhibitor I family. The properties of the iso inhibitors suggest that they may play a defensive role in the latex against pathogens and/or herbivores.

**Keywords:** *Hevea brasiliensis*; Euphorbiaceae; Proteinase iso inhibitors; Wounding; Plant defense

Gareth Norton, Arokiaraj Pappusamy, Faridah Yusof, Valerie Pujade-Renaud, Mark Perkins, David Griffiths, Heddwyn Jones, Characterisation of recombinant *Hevea brasiliensis* allene oxide synthase: Effects of cyclooxygenase inhibitors, lipoxygenase inhibitors and salicylates on enzyme activity,

**Plant Physiology and Biochemistry**, Volume 45, Issue 2, February 2007, Pages 129-138, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2007.01.003.

(<http://www.sciencedirect.com/science/article/pii/S098194280700006X>)

### **Abstract:**

Mechanical wounding and jasmonic acid (JA) treatment have been shown to be important factors in controlling laticifer differentiation in *Hevea brasiliensis* (rubber tree).

With the long-term aim of potentially modifying the endogenous levels of JA in *H. brasiliensis* by gene transfer, we describe in this paper the molecular cloning of a *H. brasiliensis* allene oxide synthase (AOS) cDNA and biochemical characterisation of the recombinant AOS (His6-HbAOS) enzyme. The AOS cDNA encodes a protein with the expected motifs present in CYP74A sub-group of the cytochrome P450 super-family of enzymes that metabolise 13-hydroperoxylinolenic acid (13-HPOT), the intermediate involved in JA synthesis. The recombinant *H. brasiliensis* AOS enzyme was estimated to have a high binding affinity for 13-HPOT with a  $K_m$  value of  $4.02 \pm 0.64$  [ $\mu$ ]M. Consistent with previous studies, mammalian cyclooxygenase (COX) and lipoxygenase (LOX) inhibitors were shown to significantly reduce His6-HbAOS enzyme activity. Although JA had no effect on His6-HbAOS, salicylic acid (SA) was shown to significantly inhibit the recombinant AOS enzyme activity in a dose dependent manner. Moreover, it was demonstrated that SA, and various analogues of SA, acted as competitive inhibitors of His6-HbAOS when 13-HPOT was used as substrate. We speculate that this effect of salicylates on AOS activity may be important in cross-talking between the SA and JA signalling pathways in plants during biotic/abiotic stress.

**Keywords: Allene oxide synthase; Hevea brasiliensis; Jasmonic acid; Laticifers**

Wilaiwan Chotigeat, Sarapee Duangchu, Rapepun Wititsuwannakun, Amornrat Phongdara, Cloning and characterization of pectate lyase from *Hevea brasiliensis*, *Plant Physiology and Biochemistry*, Volume 47, Issue 4, April 2009, Pages 243-247, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2008.12.008.

(<http://www.sciencedirect.com/science/article/pii/S0981942808002374>)

**Abstract:**

Latex from the commercial *Hevea brasiliensis* contains 30-50% (w/w) of natural rubber (cis-1,4-polyisoprene), the raw material for the many products of the rubber industry. We have constructed a cDNA library from the latex of *H. brasiliensis* to investigate the expressed genes and molecular events in the latex. We have isolated two cDNAs from this library, Hb-PEL-1 and Hb-PEL-2 that could encode for pectate lyase enzymes (EC4.2.2.2). From their sequence analysis Hb-PEL-1 and Hb-PEL-2 encode for proteins of 393 and 323 amino acids, respectively. Comparison of these deduced amino acid sequences with other pectate lyase enzymes showed they contained the conserved NADPH, Ca<sup>2+</sup> and substrate binding sites and had a 74% identity to *Arabidopsis thaliana* pectate lyase. Only the Hb-PEL-1 recombinant protein expressed from *Escherichia coli* had enzymic activity which was Ca<sup>2+</sup> dependent. Interestingly, Hb-PEL-1 contained an extra internal peptide between amino acid residue 38-108 when compared to Hb-PEL-2 and this peptide was also present in other pectate lyase enzymes. The transcript of pectate lyase (Hb-PEL) in the latex of rubber tree at various times after the first tapping was quantified by real-time PCR using 18s genes as internal standard. Most transcripts were detected on the first day after tapping and then decreased with time. This indicates that the pectate lyase may be involved in either the release of latex by breaking down the laticifer wall or in the development of laticifers.

**Keywords: cDNA library; Hevea brasiliensis; Latex; Pectate lyase; Rubber tree**

Jiahong Zhu, Quanqi Zhang, Rui Wu, Zhili Zhang, HbMT2, an ethephon-induced metallothionein gene from *Hevea brasiliensis* responds to H<sub>2</sub>O<sub>2</sub> stress, *Plant Physiology and Biochemistry*, Volume 48, Issue 8, August 2010, Pages 710-715, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2010.04.004.

(<http://www.sciencedirect.com/science/article/pii/S0981942810000823>)

**Abstract:**

Metallothioneins (MTs) are the cysteine-rich proteins with low molecular weight, which play important roles in maintaining intracellular ion homeostasis, detoxification of heavy metal ions and protecting against intracellular oxidative damages. In this study a novel ethephon-induced metallothionein gene, designated as HbMT2, was isolated and characterized from *Hevea brasiliensis*. The HbMT2 cDNA contained a 237 bp open reading frame encoding 78 amino acids and the deduced protein showed high similarity to the type 2 MTs from other plant species. Expression analysis revealed more significant accumulation of HbMT2 transcripts in leaves and latex than in roots and barks. The transcription of HbMT2 in latex was strongly induced by ethephon and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) stress. Overproduction of recombinant HbMT2 protein gave the *Escherichia coli* cells more tolerance on Cu<sup>2+</sup> and Zn<sup>2+</sup>, and the recombinant HbMT2 could scavenge the reactive oxidant species (ROS) in vitro. All these results indicated that HbMT2 could respond to ethephon stimulation and H<sub>2</sub>O<sub>2</sub> stress as a ROS scavenger in *H. brasiliensis*. It is also suggested that HbMT2 function in improving the tolerance of rubber trees to heavy metal ions, and repressing the ethephon-induced senilism and tapping panel dryness (TPD) development by ROS scavenge system in *H. brasiliensis*.

**Keywords:** *Hevea brasiliensis*; Metallothionein; Ethephon; ROS; H<sub>2</sub>O<sub>2</sub>

Rapepun Wititsuwannakul, Piyaporn Pasitkul, Kamonwan Kanokwiroon, Dhirayos Wititsuwannakul, A role for a *Hevea* latex lectin-like protein in mediating rubber particle aggregation and latex coagulation,

*Phytochemistry*, Volume 69, Issue 2, January 2008, Pages 339-347, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2007.08.019.

(<http://www.sciencedirect.com/science/article/pii/S0031942207005274>)

**Abstract:**

An in vitro aggregation of washed lutoid membrane and rubber particles, respectively, prepared from the bottom (lutoid) fraction and rubber layer of centrifuged fresh latex, leading to the formation of rubber coagulum necessary for a latex coagulation was demonstrated. A Triton X-100 extract of washed lutoid membrane proteins, isolated and prepared from the bottom fraction of centrifuged fresh latex was examined for its role in the latex coagulation process. It induced agglutination of rabbit erythrocytes, indicating the presence of a lectin-like protein. *Hevea* latex lectin-like protein (HLL) was purified to homogeneity by active chitin binding separation, followed by DEAE-Sepharose chromatography. Its Mr analyzed by SDS-PAGE was 17 kDa, whereas that determined by gel filtration was 267 kDa. The HLL had a pI value of 7.2. Several glycoproteins were shown to inhibit the HLL-induced hemagglutination. The hemagglutinin activity of HLL was enhanced by Ca<sup>2+</sup>. Of most interest was the finding that HLL strongly induced aggregation of the *Hevea* latex rubber particles (RP). This strong RP aggregation leads

to latex coagulation, indicating the possibility that it is involved in the formation of the coagulum that plugs the latex vessel ends and stops the flow of latex upon tapping. In addition, the purified HLL also induced aggregation of RP taken from several other non-Hevea latex producing plants. This might indicate either a common or universal role of this lectin-like protein in RP aggregation and hence latex coagulation. This paper, for the first time, provides clear and unequivocal evidence for either a key biological role or physiological function of an endogeneous latex lectin-like protein in the sequential process of latex coagulation.

**Keywords:** *Hevea brasiliensis*; Euphorbiaceae; Rubber latex; Lectin; Agglutinin; Rubber particle; Latex coagulation

Rapepun Wititsuwannakul, Kamonchanok Rukseree, Kamonwan Kanokwiroon, Dhirayos Wititsuwannakul, A rubber particle protein specific for Hevea latex lectin binding involved in latex coagulation,

*Phytochemistry*, Volume 69, Issue 5, March 2008, Pages 1111-1118, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2007.12.007.

(<http://www.sciencedirect.com/science/article/pii/S0031942207007236>)

**Abstract:**

In the first of this three paper series, an in vitro latex coagulation was shown to arise from aggregation of rubber particles (RP) and luteoid membranes. RP aggregation was shown to be induced by a specific Hevea latex lectin-like protein (HLL) present on the luteoid membrane. In this second paper, a binding protein (BP) ligand counterpart for HLL was identified. This RP-HLLBP, having a specific interaction, with HLL was isolated from RP and characterized. The protein was extracted from the small RP in the presence of a surfactant (0.2% Triton-X-100) and further purified to homogeneity. Purification steps included acetone precipitation, heat-treatment, and column chromatography. The presence of RP-HLLBP was monitored by its ability to compete with erythrocytes in the hemagglutination inhibition (HI) assay. The purified RP-HLLBP had an HI titre of 1.37 [ $\mu$ ]g ml<sup>-1</sup>, a pI value of 5.4, optimum activity at pH 5-8 and was thermostable up to 60 °C. On SDS-PAGE a single glycoprotein with Mr of 24 kDa was detected while on native PAGE the major Mr was about 120 kDa. The purified RP-HLLBP was shown to inhibit latex coagulation. Chitinase, but no other glycosidase tested, abolished its HI action and inhibited HLL-induced RP aggregation in a competitive dose dependent manner. This indicated the presence of, and role for, N-acetylglucosamine residues in the binding recognition. The Hevea latex lectin-like protein can thus be referred to as a Hevea latex lectin. Based on protein identification by peptide mass fingerprinting, the RP-HLLBP was confirmed to be the small rubber particle protein (SRPP). This work has unambiguously determined the role of an intrinsic RP glycoprotein (RP-HLLBP or SRPP) as a key component in formation of the rubber latex coagulum.

**Keywords:** *Hevea brasiliensis*; Euphorbiaceae; Rubber particle; Lectin receptor; Rubber particle protein; Rubber latex; Latex coagulation



K.M. Hosamani, K.S. Katagi, Characterization and structure elucidation of 12-hydroxyoctadec-cis-9-enoic acid in *Jatropha gossypifolia* and *Hevea brasiliensis* seed oils: a rich source of hydroxy fatty acid,

***Chemistry and Physics of Lipids***, Volume 152, Issue 1, March 2008, Pages 9-12, ISSN 0009-3084, DOI: 10.1016/j.chemphyslip.2007.11.003.

(<http://www.sciencedirect.com/science/article/pii/S0009308407004628>)

**Abstract:**

The seed oils containing hydroxy fatty acids are being used in various industries such as, in the production of nylon-11, in the manufacture of multi-purpose greases, as good anti-rust, new additives in water-soluble cutting fluids, and antimicrobial activity of various derivatives of ricinoleic acid polymers. *Jatropha gossypifolia* and *Hevea brasiliensis* seed oils were found to contain 18.5% and 18.0% of 12-hydroxyoctadec-cis-9-enoic acid (ricinoleic acid), respectively. The identification and characterization was based on UV, FTIR, <sup>1</sup>H NMR, <sup>13</sup>C NMR, MS, GC analysis and chemical degradations.

**Keywords:** *Jatropha gossypifolia*; *Hevea brasiliensis*; Seed oils; Fatty acids; Unusual fatty acid; 12-Hydroxyoctadec-cis-9-enoic acid; Industrial utilization

Zhi-Li Zhang, Jia-Hong Zhu, Quan-Qi Zhang, Yuan-Bao Cai, Molecular characterization of an ethephon-induced Hsp70 involved in high and low-temperature responses in *Hevea brasiliensis*,

***Plant Physiology and Biochemistry***, Volume 47, Issue 10, October 2009, Pages 954-959, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2009.06.003.

(<http://www.sciencedirect.com/science/article/pii/S0981942809001454>)

**Abstract:**

Hsp70s have been shown to play important roles in helping cells to cope with adverse environments, especially in response to temperature. In this study a novel ethephon-induced Hsp gene, designated as HbHsp70, was isolated from *Hevea brasiliensis*. The HbHsp70 cDNA contained a 1965 bp open reading frame encoding 655 amino acids. The deduced HbHsp70 protein showed high identities to Hsp70s from other plants. Expression studies revealed more significant accumulation of HbHsp70 transcripts in leaves and stems than in roots, barks and latex. The transcription of HbHsp70 was induced by ethephon, heat treatment and low temperature stress, whereas jasmonic acid had little effects. Recombinant HbHsp70 was expressed in *Escherichia coli* and purified by Ni-NTA affinity chromatography. Measuring the light scattering of luciferase (Luc) revealed that HbHsp70 prevents the aggregation of luc during high-temperature stress. In vitro experiments showed that HbHsp70 had protective functions not only against heat stress but also against chilling stress. All these data suggest that HbHsp70 may play roles in responses to heat shock and low temperature in *H. brasiliensis*.

**Keywords:** *Hevea brasiliensis*; Heat shock protein 70; Ethephon; Expression; Low temperature

Rapepun Wititsuwannakul, Piyaporn Pasitkul, Pattavuth Jewtragoon, Dhirayos Wititsuwannakul, Hevea latex lectin binding protein in C-serum as an anti-latex coagulating factor and its role in a proposed new model for latex coagulation, *Phytochemistry*, Volume 69, Issue 3, February 2008, Pages 656-662, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2007.09.021.

(<http://www.sciencedirect.com/science/article/pii/S0031942207005936>)

**Abstract:**

A distinct protein specifically recognized by its strong interaction with Hevea latex lectin (HLL) was detected in the aqueous C-serum fraction of centrifuged fresh latex. This C-serum lectin binding protein (CS-HLLBP) exhibited strong inhibition of HLL-induced hemagglutination. The CS-HLLBP was purified to homogeneity by a protocol that included ammonium sulfate fractionation, size exclusion and ion exchange chromatography. The purified CS-HLLBP had a specific HI titer of 0.23 [ $\mu$ ]g ml<sup>-1</sup>. Its Mr analyzed by SDS-PAGE was ca. 40 kDa and that by gel filtration was ca. 204 kDa. It has a pI value of 4.7, an optimum activity between pH 6 and 10 and was heat stable up to 50 [degree sign]C. The HI activity of CS-HLLBP was abolished upon treatment with chitinase. The CS-HLLBP inhibited HLL-induced rubber particle aggregation in a dose dependent manner. A highly positive correlation between CS-HLLBP activity and rubber yield per tapping was found. The correlations for fresh latex ( $r = 0.98$ ,  $P < 0.01$ ) and dry rubber ( $r = 0.95$ ,  $P < 0.01$ ) were both highly significant. This indicated that the CS-HLLBP might be used as a reliable marker for the mass screening of young seedlings to identify and select clones with potential to be superior producers of rubber. A latex anti-coagulating role of the CS-HLLBP is proposed. The findings described in this 3 paper series have been used to propose a new model of rubber latex coagulation that logically describes roles for the newly characterized latex lectin and the two lectin binding proteins.

**Keywords:** Hevea brasiliensis; Euphorbiaceae; Rubber latex; C-serum; Lectin; Lectin binding protein; [ $\alpha$ ]-Globulin; Latex flow; Anti-coagulating factor; Latex coagulation

Wenshuang Xie, Colleen M. McMahan, Amanda J. DeGraw, Mark D. Distefano, Katrina Cornish, Maureen C. Whalen, David K. Shintani, Initiation of rubber biosynthesis: In vitro comparisons of benzophenone-modified diphosphate analogues in three rubber-producing species,

*Phytochemistry*, Volume 69, Issue 14, October 2008, Pages 2539-2545, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2008.07.011.

(<http://www.sciencedirect.com/science/article/pii/S0031942208003531>)

**Abstract:**

Natural rubber, cis-1,4-polyisoprene, is a vital industrial material synthesized by plants via a side branch of the isoprenoid pathway by the enzyme rubber transferase. While the specific structure of this enzyme is not yet defined, based on activity it is probably a cis-prenyl transferase. Photoactive functionalized substrate analogues have been successfully used to identify isoprenoid-utilizing enzymes such as cis- and trans-prenyltransferases, and initiator binding of an allylic pyrophosphate molecule in rubber

transferase has similar features to these systems. In this paper, a series of benzophenone-modified initiator analogues were shown to successfully initiate rubber biosynthesis in vitro in enzymatically-active washed rubber particles from *Ficus elastica*, *Hevea brasiliensis* and *Parthenium argentatum*.

Rubber transferases from all three species initiated rubber biosynthesis most efficiently with farnesyl pyrophosphate. However, rubber transferase had a higher affinity for benzophenone geranyl pyrophosphate (Bz-GPP) and dimethylallyl pyrophosphate (Bz-DMAPP) analogues with ether-linkages than the corresponding GPP or DMAPP. In contrast, ester-linked Bz-DMAPP analogues were less efficient initiators than DMAPP. Thus, rubber biosynthesis depends on both the size and the structure of Bz-initiator molecules. Kinetic studies thereby inform selection of specific probes for covalent photolabeling of the initiator binding site of rubber transferase.

**Keywords:** *Parthenium argentatum*; Guayule; *Ficus elastica*; *Hevea brasiliensis*; Photoaffinity labeling; Benzophenone; Allylic pyrophosphate; Farnesyl pyrophosphate; Rubber transferase; Cis-prenyltransferase

Bernardo M.T. da Costa, Jay D. Keasling, Colleen M. McMahan, Katrina Cornish, Magnesium ion regulation of in vitro rubber biosynthesis by *Parthenium argentatum* Gray,

*Phytochemistry*, Volume 67, Issue 15, Rod Croteau Special Issue, Part 1, August 2006, Pages 1621-1628, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2006.04.010.

(<http://www.sciencedirect.com/science/article/pii/S0031942206002160>)

**Abstract:**

Natural rubber is produced by a rubber transferase (a cis-prenyltransferase). Rubber transferase uses allylic pyrophosphate to initiate the rubber molecule and isopentenyl pyrophosphate (IPP) to form the polymer. Rubber biosynthesis also requires a divalent metal cation. Understanding how molecular weight is regulated is important because high molecular weight is required for high quality rubber. We characterized the in vitro effects of Mg<sup>2+</sup> on the biosynthetic rate of rubber produced by an alternative natural rubber crop, *Parthenium argentatum* (guayule). The affinity of the rubber transferase from *P. argentatum* for IPP [middle dot] Mg was shown to depend on the Mg<sup>2+</sup> concentration in a similar fashion to the *H. brasiliensis* rubber transferase, although to a less extreme degree. Also, in vitro Mg<sup>2+</sup> concentration significantly affects rubber molecular weight of both species, but molecular weight is less sensitive to Mg<sup>2+</sup> concentration in *P. argentatum* than in *H. brasiliensis*.

**Keywords:** *Parthenium argentatum*; Compositae; *Hevea brasiliensis* Muell. Arg.; Euphorbiaceae; Regulation; Rubber transferase; cis-Prenyltransferase; Magnesium; FPP; IPP

R. Lacote, O. Gabla, S. Obouayeba, J.M. Eschbach, F. Rivano, K. Dian, E. Gohet, Long-term effect of ethylene stimulation on the yield of rubber trees is linked to latex cell biochemistry,

*Field Crops Research*, Volume 115, Issue 1, 4 January 2010, Pages 94-98, ISSN 0378-4290, DOI: 10.1016/j.fcr.2009.10.007.

(<http://www.sciencedirect.com/science/article/pii/S0378429009002743>)

**Abstract:**

Ethylene stimulation with ethephon (2-chloroethylphosphonic acid) is nowadays essential for increasing latex production in the rubber tree (*Hevea brasiliensis*): both small-scale planters and agro-industrial plantations worldwide use ethephon. Ethylene stimulation strongly influences cumulative yield and latex cell biochemistry. The purpose of this study was to characterize the long-term behaviour of the rubber tree under ethephon treatment. Over a period of 7 years in Cote d'Ivoire, West Africa, the effect of eight frequencies of ethylene stimulation on yield and latex cell biochemistry was compared in four rubber trees clones, IRCA 130, IRCA 230, GT 1, and PB 217. The ability of the trees to produce more latex under ethylene stimulation was related to the sucrose and inorganic phosphorus contents of the latex cells. For high-yield clones with low sugar content and high inorganic phosphorus content like IRCA 130, namely quick starter clones, no stimulation was necessary to improve yield. For clones like IRCA 230, with higher sugar content, eight ethylene stimulations per year was the optimum frequency to obtain the highest yield. The effect of ethylene stimulation on latex yield increase was significant in clones with high sucrose content and low inorganic phosphorus content such as PB 217. These clones, namely slow starter clones, needed more stimulation to produce more, but in the longer run there were no negative effects of ethylene on the latex yield. These results will help planters optimize latex production by choosing the most appropriate ethylene stimulation to clones according to their latex cells biochemistry.

**Keywords:** *Hevea brasiliensis*; Ethylene stimulation; Latex cell biochemistry; Sucrose; Inorganic phosphorus; Latex yield

Bradley S. Bushman, Andrew A. Scholte, Katrina Cornish, Deborah J. Scott, Jenny L. Brichta, John C. Vederas, Oswaldo Ochoa, Richard W. Michelmore, David K. Shintani, Steven J. Knapp, Identification and comparison of natural rubber from two *Lactuca* species,

*Phytochemistry*, Volume 67, Issue 23, December 2006, Pages 2590-2596, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2006.09.012.

(<http://www.sciencedirect.com/science/article/pii/S0031942206005760>)

**Abstract:**

Renewed interest in the identification of alternative sources of natural rubber to *Hevea brasiliensis* has focused on the Compositae family. In our search for Compositae models for rubber synthesis, we extracted latex from stems of two lettuce species: *Lactuca serriola*, prickly lettuce, and *Lactuca sativa* cv. Salinas, crisphead lettuce. Both species contained cis-1,4-polyisoprene rubber in the dichloromethane-soluble portions of their latex, and sesquiterpene lactones in their acetone-soluble portions. The rubber from both species and their progeny had molecular weights in excess of 1,000,000 g/mol, and polydispersity values of 1.1. Rubber transferase activity was detected across a range of farnesyl diphosphate initiator concentrations, with decreased activity as initiator concentrations exceeded putative saturation. These results add lettuce to the short list of plant species that produce high molecular weight rubber in their latex. Due

to the genomic and agronomic resources available in lettuce species, they provide the opportunity for further dissection of natural rubber biosynthesis in plants.

**Keywords:** *Lactuca sativa*; *Lactuca serriola*; *Compositae*; Rubber; cis-1,4-Polyisoprene; *Lactuca*; Lettuce

Cesar A. Reyes-Lopez, Martha Pedraza-Escalona, Guillermo Mendoza, Alejandra Hernandez-Santoyo, Adela Rodriguez-Romero, A single amino acid substitution on the surface of a natural hevein isoform (Hev b 6.0202), confers different IgE recognition, *FEBS Letters*, Volume 580, Issue 10, 1 May 2006, Pages 2483-2487, ISSN 0014-5793, DOI: 10.1016/j.febslet.2006.03.085.

(<http://www.sciencedirect.com/science/article/pii/S0014579306004121>)

**Abstract:**

Decreased immune reactivity of isoforms of major allergens has been reported. However, such claims have always been based on experiments with recombinant proteins. This work describes the molecular and physicochemical characterization of a hevein (Hev b 6.0201) natural isoform (Hev b 6.0202), which is present in rubber latex from *Hevea brasiliensis*. The isoallergen has a single substitution Asn14Asp, which gives rise to local differences in the surface potential, as observed from the crystal structure presented here. Besides, ELISA inhibition using serum pools of adult and pediatric patients showed reduced IgE-binding capacity (~27%) with the isoallergen. Overall, these results are relevant to delineate crucial residues involved in this dominant discontinuous epitope.

**Keywords:** Crystal structure; Isoallergens; Hevein; Hev b 6.0201; IgE-epitope

C.R. Benedict, Paul J. Greer, M.A. Foster, The physiological and biochemical responses of guayule to the low temperature of the Chihuahuan Desert in the biosynthesis of rubber,

*Industrial Crops and Products*, Volume 27, Issue 3, May 2008, Pages 225-235, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2007.09.003.

(<http://www.sciencedirect.com/science/article/pii/S0926669007001379>)

**Abstract:**

Guayule (*Parthenium argentatum* Gray) is a rubber plant indigenous to the Chihuahuan Desert of Northern Mexico and Southwestern Texas. In this review we report the nature of the physiological, cellular and biochemical responses of these plants to the low temperature of the desert in the biosynthesis of rubber.

Studies on rubber formation in guayule in several field plantings support the conclusion that the low temperature of the fall and winter months of the Chihuahuan Desert promotes a rapid increase in rubber biosynthesis. There are definite changes in the cortical parenchyma during the biosynthesis and deposition of rubber. Discrete rubber particles are formed in the parietal and interior cytosol. Following the digestion of the cytosol in the interior of the cell and rubber particle fusion leaves the parenchyma with rubber deposits throughout the cells. The rubber transferase (RT) bound to the washed rubber particles (WRP-RT) catalyzes the formation of different length cis-1, 4-polyisoprene chains that collectively form the rubber polymer. The activities of the WRP-

RT and the endoplasmic reticulum 3-hydroxy-3-methylglutaryl-coenzyme A reductase (HMGR) increase with exposure of the plant to low temperature and it is conceivable that these two enzymes play a pivotal role in the biosynthesis of rubber during this period. Thus guayule plants indigenous to the Chihuahuan Desert have developed a genetic system capable of responding to the low temperatures of the fall and winter of the desert culminating in the formation of rubber.

**Keywords: Guayule; Parthenium argentatum; Natural rubber; Rubber biosynthesis; Chihuahuan Desert**

Wannapa Sritanyarat, Gregory Pearce, William F. Siems, Clarence A. Ryan, Rapepun Wititsuwannakul, Dhirayos Wititsuwannakul, Isolation and characterization of iso inhibitors of the potato protease inhibitor I family from the latex of the rubber trees, *Hevea brasiliensis*,

*Phytochemistry*, Volume 67, Issue 15, Rod Croteau Special Issue, Part 1, August 2006, Pages 1644-1650, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2005.12.016. (<http://www.sciencedirect.com/science/article/pii/S0031942205006795>)

**Abstract:**

Three iso inhibitors have been isolated to homogeneity from the C-serum of the latex of the rubber tree, *Hevea brasiliensis* clone RRIM 600, and named HPI-1, HPI-2a and HPI-2b. The three inhibitors share the same amino acid sequence (69 residues) but the masses of the three forms were determined to be 14,893 +/- 10, 7757 +/- 5, and 7565 +/- 5, respectively, indicating that post-translational modifications of the protein have occurred during latex collection. One adduct could be removed by reducing agents, and was determined to be glutathione, while the other adduct could not be removed by reducing agents and has not been identified. The N-termini of the inhibitor proteins were blocked by an acetylated Ala, but the complete amino acid sequence analysis of the deblocked inhibitors by Edman degradation of fragments from endopeptidase C digestion and mass spectrometry confirmed that the three iso inhibitors were derived from a single protein. The amino acid sequence of the protein differed at two positions from the sequence deduced from a cDNA reported in GenBank. The gene coding for the inhibitor is wound-inducible and is a member of the potato inhibitor I family of protease inhibitors. The inhibitor strongly inhibited subtilisin A, weakly inhibited trypsin, and did not inhibit chymotrypsin. The amino acid residues at the reactive site P1 and were determined to be Gln45 and Asp46, respectively, residues rarely reported at the reactive site in potato inhibitor I family members. Comparison of amino acid sequences revealed that the HPI iso inhibitors shared from 33% to 55% identity (50-74% similarity) to inhibitors of the potato inhibitor I family. The properties of the iso inhibitors suggest that they may play a defensive role in the latex against pathogens and/or herbivores.

**Keywords: Hevea brasiliensis; Euphorbiaceae; Proteinase iso inhibitors; Wounding; Plant defense**

Jiahong Zhu, Quanqi Zhang, Rui Wu, Zhili Zhang, HbMT2, an ethephon-induced metallothionein gene from *Hevea brasiliensis* responds to H<sub>2</sub>O<sub>2</sub> stress, *Plant Physiology and Biochemistry*, Volume 48, Issue 8, August 2010, Pages 710-715, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2010.04.004. (<http://www.sciencedirect.com/science/article/pii/S0981942810000823>)

**Abstract:**

Metallothioneins (MTs) are the cysteine-rich proteins with low molecular weight, which play important roles in maintaining intracellular ion homeostasis, detoxification of heavy metal ions and protecting against intracellular oxidative damages. In this study a novel ethephon-induced metallothionein gene, designated as HbMT2, was isolated and characterized from *Hevea brasiliensis*. The HbMT2 cDNA contained a 237 bp open reading frame encoding 78 amino acids and the deduced protein showed high similarity to the type 2 MTs from other plant species. Expression analysis revealed more significant accumulation of HbMT2 transcripts in leaves and latex than in roots and barks. The transcription of HbMT2 in latex was strongly induced by ethephon and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) stress. Overproduction of recombinant HbMT2 protein gave the *Escherichia coli* cells more tolerance on Cu<sup>2+</sup> and Zn<sup>2+</sup>, and the recombinant HbMT2 could scavenge the reactive oxidant species (ROS) in vitro. All these results indicated that HbMT2 could respond to ethephon stimulation and H<sub>2</sub>O<sub>2</sub> stress as a ROS scavenger in *H. brasiliensis*. It is also suggested that HbMT2 function in improving the tolerance of rubber trees to heavy metal ions, and repressing the ethephon-induced senilism and tapping panel dryness (TPD) development by ROS scavenge system in *H. brasiliensis*.

**Keywords:** *Hevea brasiliensis*; Metallothionein; Ethephon; ROS; H<sub>2</sub>O<sub>2</sub>

Gareth Norton, Arokia Raj Pappusamy, Faridah Yusof, Valerie Pujade-Renaud, Mark Perkins, David Griffiths, Heddwyn Jones, Characterisation of recombinant *Hevea brasiliensis* allene oxide synthase: Effects of cyclooxygenase inhibitors, lipoxygenase inhibitors and salicylates on enzyme activity, *Plant Physiology and Biochemistry*, Volume 45, Issue 2, February 2007, Pages 129-138, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2007.01.003. (<http://www.sciencedirect.com/science/article/pii/S098194280700006X>)

**Abstract:**

Mechanical wounding and jasmonic acid (JA) treatment have been shown to be important factors in controlling laticifer differentiation in *Hevea brasiliensis* (rubber tree). With the long-term aim of potentially modifying the endogenous levels of JA in *H. brasiliensis* by gene transfer, we describe in this paper the molecular cloning of a *H. brasiliensis* allene oxide synthase (AOS) cDNA and biochemical characterisation of the recombinant AOS (His6-HbAOS) enzyme. The AOS cDNA encodes a protein with the expected motifs present in CYP74A sub-group of the cytochrome P450 super-family of enzymes that metabolise 13-hydroperoxylinolenic acid (13-HPOT), the intermediate involved in JA synthesis. The recombinant *H. brasiliensis* AOS enzyme was estimated to have a high binding affinity for 13-HPOT with a K<sub>m</sub> value of 4.02 ± 0.64 [μM]. Consistent with previous studies, mammalian cyclooxygenase (COX) and lipoxygenase (LOX) inhibitors were shown to significantly reduce His6-HbAOS enzyme activity.

Although JA had no effect on His6-HbAOS, salicylic acid (SA) was shown to significantly inhibit the recombinant AOS enzyme activity in a dose dependent manner. Moreover, it was demonstrated that SA, and various analogues of SA, acted as competitive inhibitors of His6-HbAOS when 13-HPOT was used as substrate. We speculate that this effect of salicylates on AOS activity may be important in cross-talking between the SA and JA signalling pathways in plants during biotic/abiotic stress.

**Keywords: Allene oxide synthase; Hevea brasiliensis; Jasmonic acid; Laticifers**

K.M. Hosamani, K.S. Katagi, Characterization and structure elucidation of 12-hydroxyoctadec-cis-9-enoic acid in *Jatropha gossypifolia* and *Hevea brasiliensis* seed oils: a rich source of hydroxy fatty acid,

***Chemistry and Physics of Lipids***, Volume 152, Issue 1, March 2008, Pages 9-12, ISSN 0009-3084, DOI: 10.1016/j.chemphyslip.2007.11.003.

(<http://www.sciencedirect.com/science/article/pii/S0009308407004628>)

**Abstract:**

The seed oils containing hydroxy fatty acids are being used in various industries such as, in the production of nylon-11, in the manufacture of multi-purpose greases, as good anti-rust, new additives in water-soluble cutting fluids, and antimicrobial activity of various derivatives of ricinoleic acid polymers. *Jatropha gossypifolia* and *Hevea brasiliensis* seed oils were found to contain 18.5% and 18.0% of 12-hydroxyoctadec-cis-9-enoic acid (ricinoleic acid), respectively. The identification and characterization was based on UV, FTIR, <sup>1</sup>H NMR, <sup>13</sup>C NMR, MS, GC analysis and chemical degradations.

**Keywords: *Jatropha gossypifolia*; *Hevea brasiliensis*; Seed oils; Fatty acids; Unusual fatty acid; 12-Hydroxyoctadec-cis-9-enoic acid; Industrial utilization**

Rapepun Wititsuwannakul, Piyaporn Pasitkul, Pattavuth Jewtragoon, Dhirayos Wititsuwannakul, Hevea latex lectin binding protein in C-serum as an anti-latex coagulating factor and its role in a proposed new model for latex coagulation, ***Phytochemistry***, Volume 69, Issue 3, February 2008, Pages 656-662, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2007.09.021.

(<http://www.sciencedirect.com/science/article/pii/S0031942207005936>)

**Abstract:**

A distinct protein specifically recognized by its strong interaction with Hevea latex lectin (HLL) was detected in the aqueous C-serum fraction of centrifuged fresh latex. This C-serum lectin binding protein (CS-HLLBP) exhibited strong inhibition of HLL-induced hemagglutination. The CS-HLLBP was purified to homogeneity by a protocol that included ammonium sulfate fractionation, size exclusion and ion exchange chromatography. The purified CS-HLLBP had a specific HI titer of 0.23 [ $\mu$ ]g ml<sup>-1</sup>. Its Mr analyzed by SDS-PAGE was ca. 40 kDa and that by gel filtration was ca. 204 kDa. It has a pI value of 4.7, an optimum activity between pH 6 and 10 and was heat stable up to 50 [degree sign]C. The HI activity of CS-HLLBP was abolished upon treatment with chitinase. The CS-HLLBP inhibited HLL-induced rubber particle aggregation in a dose dependent manner. A highly positive correlation between CS-HLLBP activity and



rubber yield per tapping was found. The correlations for fresh latex ( $r = 0.98$ ,  $P < 0.01$ ) and dry rubber ( $r = 0.95$ ,  $P < 0.01$ ) were both highly significant. This indicated that the CS-HLLBP might be used as a reliable marker for the mass screening of young seedlings to identify and select clones with potential to be superior producers of rubber. A latex anti-coagulating role of the CS-HLLBP is proposed. The findings described in this 3 paper series have been used to propose a new model of rubber latex coagulation that logically describes roles for the newly characterized latex lectin and the two lectin binding proteins.

**Keywords:** *Hevea brasiliensis*; Euphorbiaceae; Rubber latex; C-serum; Lectin; Lectin binding protein; [ $\alpha$ ]-Globulin; Latex flow; Anti-coagulating factor; Latex coagulation

Wenshuang Xie, Colleen M. McMahan, Amanda J. DeGraw, Mark D. Distefano, Katrina Cornish, Maureen C. Whalen, David K. Shintani, Initiation of rubber biosynthesis: In vitro comparisons of benzophenone-modified diphosphate analogues in three rubber-producing species,

*Phytochemistry*, Volume 69, Issue 14, October 2008, Pages 2539-2545, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2008.07.011.

(<http://www.sciencedirect.com/science/article/pii/S0031942208003531>)

**Abstract:**

Natural rubber, cis-1,4-polyisoprene, is a vital industrial material synthesized by plants via a side branch of the isoprenoid pathway by the enzyme rubber transferase. While the specific structure of this enzyme is not yet defined, based on activity it is probably a cis-prenyl transferase. Photoactive functionalized substrate analogues have been successfully used to identify isoprenoid-utilizing enzymes such as cis- and trans-prenyltransferases, and initiator binding of an allylic pyrophosphate molecule in rubber transferase has similar features to these systems. In this paper, a series of benzophenone-modified initiator analogues were shown to successfully initiate rubber biosynthesis in vitro in enzymatically-active washed rubber particles from *Ficus elastica*, *Hevea brasiliensis* and *Parthenium argentatum*. Rubber transferases from all three species initiated rubber biosynthesis most efficiently with farnesyl pyrophosphate. However, rubber transferase had a higher affinity for benzophenone geranyl pyrophosphate (Bz-GPP) and dimethylallyl pyrophosphate (Bz-DMAPP) analogues with ether-linkages than the corresponding GPP or DMAPP. In contrast, ester-linked Bz-DMAPP analogues were less efficient initiators than DMAPP. Thus, rubber biosynthesis depends on both the size and the structure of Bz-initiator molecules. Kinetic studies thereby inform selection of specific probes for covalent photolabeling of the initiator binding site of rubber transferase.

**Keywords:** *Parthenium argentatum*; Guayule; *Ficus elastica*; *Hevea brasiliensis*; Photoaffinity labeling; Benzophenone; Allylic pyrophosphate; Farnesyl pyrophosphate; Rubber transferase; Cis-prenyltransferase

Bernardo M.T. da Costa, Jay D. Keasling, Colleen M. McMahan, Katrina Cornish, Magnesium ion regulation of in vitro rubber biosynthesis by *Parthenium argentatum* Gray,

**Phytochemistry**, Volume 67, Issue 15, Rod Croteau Special Issue, Part 1, August 2006, Pages 1621-1628, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2006.04.010. (<http://www.sciencedirect.com/science/article/pii/S0031942206002160>)

**Abstract:**

Natural rubber is produced by a rubber transferase (a cis-prenyltransferase). Rubber transferase uses allylic pyrophosphate to initiate the rubber molecule and isopentenyl pyrophosphate (IPP) to form the polymer. Rubber biosynthesis also requires a divalent metal cation. Understanding how molecular weight is regulated is important because high molecular weight is required for high quality rubber. We characterized the in vitro effects of Mg<sup>2+</sup> on the biosynthetic rate of rubber produced by an alternative natural rubber crop, *Parthenium argentatum* (guayule). The affinity of the rubber transferase from *P. argentatum* for IPP [middle dot] Mg was shown to depend on the Mg<sup>2+</sup> concentration in a similar fashion to the *H. brasiliensis* rubber transferase, although to a less extreme degree. Also, in vitro Mg<sup>2+</sup> concentration significantly affects rubber molecular weight of both species, but molecular weight is less sensitive to Mg<sup>2+</sup> concentration in *P. argentatum* than in *H. brasiliensis*.

**Keywords:** *Parthenium argentatum*; *Compositae*; *Hevea brasiliensis* Muell. Arg.; *Euphorbiaceae*; Regulation; Rubber transferase; cis-Prenyltransferase; Magnesium; FPP; IPP

R. Lacote, O. Gabla, S. Obouayeba, J.M. Eschbach, F. Rivano, K. Dian, E. Gohet, Long-term effect of ethylene stimulation on the yield of rubber trees is linked to latex cell biochemistry,

**Field Crops Research**, Volume 115, Issue 1, 4 January 2010, Pages 94-98, ISSN 0378-4290, DOI: 10.1016/j.fcr.2009.10.007.

(<http://www.sciencedirect.com/science/article/pii/S0378429009002743>)

**Abstract:**

Ethylene stimulation with ethephon (2-chloroethylphosphonic acid) is nowadays essential for increasing latex production in the rubber tree (*Hevea brasiliensis*): both small-scale planters and agro-industrial plantations worldwide use ethephon. Ethylene stimulation strongly influences cumulative yield and latex cell biochemistry. The purpose of this study was to characterize the long-term behaviour of the rubber tree under ethephon treatment. Over a period of 7 years in Cote d'Ivoire, West Africa, the effect of eight frequencies of ethylene stimulation on yield and latex cell biochemistry was compared in four rubber trees clones, IRCA 130, IRCA 230, GT 1, and PB 217. The ability of the trees to produce more latex under ethylene stimulation was related to the sucrose and inorganic phosphorus contents of the latex cells. For high-yield clones with low sugar content and high inorganic phosphorus content like IRCA 130, namely quick starter clones, no stimulation was necessary to improve yield. For clones like IRCA 230, with higher sugar content, eight ethylene stimulations per year was the optimum frequency to obtain the highest yield. The effect of ethylene stimulation on latex yield increase was significant in clones with high sucrose content and low inorganic

phosphorus content such as PB 217. These clones, namely slow starter clones, needed more stimulation to produce more, but in the longer run there were no negative effects of ethylene on the latex yield. These results will help planters optimize latex production by choosing the most appropriate ethylene stimulation to clones according to their latex cells biochemistry.

**Keywords: Hevea brasiliensis; Ethylene stimulation; Latex cell biochemistry; Sucrose; Inorganic phosphorus; Latex yield**

V.H. Teetor, D.T. Ray, W.W. Schloman Jr., Evaluating chemical indices of guayule rubber content: Guayulins A and B,

***Industrial Crops and Products***, Volume 29, Issues 2-3, March 2009, Pages 590-598, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2008.11.005.

(<http://www.sciencedirect.com/science/article/pii/S0926669008002094>)

**Abstract:**

Guayule (*Parthenium argentatum* Gray) is now a commercial crop for the production of high-quality, hypoallergenic natural rubber latex. Because guayule is relatively resistant to both insect and disease pests, its cultivation requires little chemical input. It has been postulated that guayule's chemical defense system is based on terpene derivatives such as guayulins A and B. The goals of this research were to: (1) describe the distribution of guayulins A and B throughout the plant, (2) determine whether guayulins can be used as a predictor of rubber content/yield (is guayulin content related to rubber content), and (3) determine whether the guayulin content/distribution has been changed through plant breeding. Two plants each of three different lines (11591, AZ-1, and AZ-3) and two ages (1 and 2 years old) were harvested at the soil line in the fall of 2002 and spring of 2003. Plants were separated into eight parts: brown leaves, green leaves, stem tips, stems less than 5 mm in diameter, stems between 5 and 10 mm, stems greater than 10 mm, green stems, and flower parts. Samples were analyzed for guayulins A and B and rubber content. Guayulins A and B and rubber were found in all plant parts, but were most prevalent in stems larger than 10 mm in diameter, and were significantly correlated with each other in these stems. Guayulin A was found in greater concentrations than guayulin B. There were no significant differences between plant ages for rubber, or guayulin content at the time of harvest, or guayulins for harvest season. Rubber percent was higher in spring, which agrees with numerous other investigations. Guayulin A in the stems is correlated with the total rubber in the plant, suggesting it as a potential selection tool. However, rubber in the same plant parts has a higher correlation with total plant rubber and remains a better and easier selection criterion than guayulin A. The extent to which guayulin contents differed between older, relatively unimproved lines and newer improved lines was evaluated. Variety 11591 had a higher concentration of guayulin A than guayulin B, so that the ratio of A to B was much higher than in the two improved lines (AZ-1 and AZ-3). It appears that the ratio of guayulin A to guayulin B has been changed in the two newer selections, but it is unclear whether this will affect pest resistance.

**Keywords: Guayule; Latex; Parthenium argentatum; cis-1,4-Polyisoprene; Rubber; Guayulin A; Guayulin B**

C.R. Benedict, Paul J. Greer, M.A. Foster, The physiological and biochemical responses of guayule to the low temperature of the Chihuahuan Desert in the biosynthesis of rubber,

***Industrial Crops and Products***, Volume 27, Issue 3, May 2008, Pages 225-235, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2007.09.003.

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**Abstract:**

Guayule (*Parthenium argentatum* Gray) is a rubber plant indigenous to the Chihuahuan Desert of Northern Mexico and Southwestern Texas. In this review we report the nature of the physiological, cellular and biochemical responses of these plants to the low temperature of the desert in the biosynthesis of rubber. Studies on rubber formation in guayule in several field plantings support the conclusion that the low temperature of the fall and winter months of the Chihuahuan Desert promotes a rapid increase in rubber biosynthesis. There are definite changes in the cortical parenchyma during the biosynthesis and deposition of rubber. Discrete rubber particles are formed in the parietal and interior cytosol. Following the digestion of the cytosol in the interior of the cell and rubber particle fusion leaves the parenchyma with rubber deposits throughout the cells. The rubber transferase (RT) bound to the washed rubber particles (WRP-RT) catalyzes the formation of different length cis-1, 4-polyisoprene chains that collectively form the rubber polymer. The activities of the WRP-RT and the endoplasmic reticulum 3-hydroxy-3-methylglutaryl-coenzyme A reductase (HMGR) increase with exposure of the plant to low temperature and it is conceivable that these two enzymes play a pivotal role in the biosynthesis of rubber during this period. Thus guayule plants indigenous to the Chihuahuan Desert have developed a genetic system capable of responding to the low temperatures of the fall and winter of the desert culminating in the formation of rubber.

**Keywords:** Guayule; *Parthenium argentatum*; Natural rubber; Rubber biosynthesis; Chihuahuan Desert

Wilaiwan Chotigeat, Sarapee Duangchu, Rapepun Wititsuwannakun, Amornrat Phongdara, Cloning and characterization of pectate lyase from *Hevea brasiliensis*, ***Plant Physiology and Biochemistry***, Volume 47, Issue 4, April 2009, Pages 243-247, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2008.12.008.

(<http://www.sciencedirect.com/science/article/pii/S0981942808002374>)

**Abstract:**

Latex from the commercial *Hevea brasiliensis* contains 30-50% (w/w) of natural rubber (cis-1,4-polyisoprene), the raw material for the many products of the rubber industry. We have constructed a cDNA library from the latex of *H. brasiliensis* to investigate the expressed genes and molecular events in the latex. We have isolated two cDNAs from this library, Hb-PEL-1 and Hb-PEL-2 that could encode for pectate lyase enzymes (EC4.2.2.2). From their sequence analysis Hb-PEL-1 and Hb-PEL-2 encode for proteins of 393 and 323 amino acids, respectively. Comparison of these deduced amino acid sequences with other pectate lyase enzymes showed they contained the conserved NADPH, Ca<sup>2+</sup> and substrate binding sites and had a 74% identity to *Arabidopsis thaliana* pectate lyase. Only the Hb-PEL-1 recombinant protein expressed from

Escherichia coli had enzymic activity which was Ca<sup>2+</sup> dependent. Interestingly, Hb-PEL-1 contained an extra internal peptide between amino acid residue 38-108 when compared to Hb-PEL-2 and this peptide was also present in other pectate lyase enzymes. The transcript of pectate lyase (Hb-PEL) in the latex of rubber tree at various times after the first tapping was quantified by real-time PCR using 18s genes as internal standard. Most transcripts were detected on the first day after tapping and then decreased with time. This indicates that the pectate lyase may be involved in either the release of latex by breaking down the laticifer wall or in the development of laticifers.

**Keywords: cDNA library; Hevea brasiliensis; Latex; Pectate lyase; Rubber tree**

Jiahong Zhu, Quanqi Zhang, Rui Wu, Zhili Zhang, HbMT2, an ethephon-induced metallothionein gene from Hevea brasiliensis responds to H<sub>2</sub>O<sub>2</sub> stress,

***Plant Physiology and Biochemistry***, Volume 48, Issue 8, August 2010, Pages 710-715, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2010.04.004.

(<http://www.sciencedirect.com/science/article/pii/S0981942810000823>)

**Abstract:**

Metallothioneins (MTs) are the cysteine-rich proteins with low molecular weight, which play important roles in maintaining intracellular ion homeostasis, detoxification of heavy metal ions and protecting against intracellular oxidative damages. In this study a novel ethephon-induced metallothionein gene, designated as HbMT2, was isolated and characterized from Hevea brasiliensis. The HbMT2 cDNA contained a 237 bp open reading frame encoding 78 amino acids and the deduced protein showed high similarity to the type 2 MTs from other plant species. Expression analysis revealed more significant accumulation of HbMT2 transcripts in leaves and latex than in roots and barks. The transcription of HbMT2 in latex was strongly induced by ethephon and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) stress. Overproduction of recombinant HbMT2 protein gave the Escherichia coli cells more tolerance on Cu<sup>2+</sup> and Zn<sup>2+</sup>, and the recombinant HbMT2 could scavenge the reactive oxidant species (ROS) in vitro. All these results indicated that HbMT2 could respond to ethephon stimulation and H<sub>2</sub>O<sub>2</sub> stress as a ROS scavenger in H. brasiliensis. It is also suggested that HbMT2 function in improving the tolerance of rubber trees to heavy metal ions, and repressing the ethephon-induced senilism and tapping panel dryness (TPD) development by ROS scavenge system in H. brasiliensis.

**Keywords: Hevea brasiliensis; Metallothionein; Ethephon; ROS; H<sub>2</sub>O<sub>2</sub>**

Gareth Norton, Arokiaraj Pappusamy, Faridah Yusof, Valerie Pujade-Renaud, Mark Perkins, David Griffiths, Heddwyn Jones, Characterisation of recombinant Hevea brasiliensis allene oxide synthase: Effects of cyclooxygenase inhibitors, lipoxygenase inhibitors and salicylates on enzyme activity,

***Plant Physiology and Biochemistry***, Volume 45, Issue 2, February 2007, Pages 129-138, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2007.01.003.

(<http://www.sciencedirect.com/science/article/pii/S098194280700006X>)

**Abstract:**

Mechanical wounding and jasmonic acid (JA) treatment have been shown to be important factors in controlling laticifer differentiation in *Hevea brasiliensis* (rubber tree). With the long-term aim of potentially modifying the endogenous levels of JA in *H. brasiliensis* by gene transfer, we describe in this paper the molecular cloning of a *H. brasiliensis* allene oxide synthase (AOS) cDNA and biochemical characterisation of the recombinant AOS (His6-HbAOS) enzyme. The AOS cDNA encodes a protein with the expected motifs present in CYP74A sub-group of the cytochrome P450 super-family of enzymes that metabolise 13-hydroperoxylinolenic acid (13-HPOT), the intermediate involved in JA synthesis. The recombinant *H. brasiliensis* AOS enzyme was estimated to have a high binding affinity for 13-HPOT with a  $K_m$  value of  $4.02 \pm 0.64$  [ $\mu$ ]M. Consistent with previous studies, mammalian cyclooxygenase (COX) and lipoxygenase (LOX) inhibitors were shown to significantly reduce His6-HbAOS enzyme activity. Although JA had no effect on His6-HbAOS, salicylic acid (SA) was shown to significantly inhibit the recombinant AOS enzyme activity in a dose dependent manner. Moreover, it was demonstrated that SA, and various analogues of SA, acted as competitive inhibitors of His6-HbAOS when 13-HPOT was used as substrate. We speculate that this effect of salicylates on AOS activity may be important in cross-talking between the SA and JA signalling pathways in plants during biotic/abiotic stress.

**Keywords:** Allene oxide synthase; *Hevea brasiliensis*; Jasmonic acid; Laticifers

Rapepun Wititsuwannakul, Piyaporn Pasitkul, Kamonwan Kanokwiroon, Dhirayos Wititsuwannakul, A role for a *Hevea* latex lectin-like protein in mediating rubber particle aggregation and latex coagulation,

*Phytochemistry*, Volume 69, Issue 2, January 2008, Pages 339-347, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2007.08.019.

(<http://www.sciencedirect.com/science/article/pii/S0031942207005274>)

**Abstract:**

An in vitro aggregation of washed luteoid membrane and rubber particles, respectively, prepared from the bottom (luteoid) fraction and rubber layer of centrifuged fresh latex, leading to the formation of rubber coagulum necessary for a latex coagulation was demonstrated. A Triton X-100 extract of washed luteoid membrane proteins, isolated and prepared from the bottom fraction of centrifuged fresh latex was examined for its role in the latex coagulation process. It induced agglutination of rabbit erythrocytes, indicating the presence of a lectin-like protein. *Hevea* latex lectin-like protein (HLL) was purified to homogeneity by active chitin binding separation, followed by DEAE-Sepharose chromatography. Its  $M_r$  analyzed by SDS-PAGE was 17 kDa, whereas that determined by gel filtration was 267 kDa. The HLL had a pI value of 7.2. Several glycoproteins were shown to inhibit the HLL-induced hemagglutination. The hemagglutinin activity of HLL was enhanced by  $Ca^{2+}$ . Of most interest was the finding that HLL strongly induced aggregation of the *Hevea* latex rubber particles (RP). This strong RP aggregation leads to latex coagulation, indicating the possibility that it is involved in the formation of the coagulum that plugs the latex vessel ends and stops the flow of latex upon tapping. In addition, the purified HLL also induced aggregation of RP taken from several other non-*Hevea* latex producing plants. This might indicate either a common or universal role of

this lectin-like protein in RP aggregation and hence latex coagulation. This paper, for the first time, provides clear and unequivocal evidence for either a key biological role or physiological function of an endogeneous latex lectin-like protein in the sequential process of latex coagulation.

**Keywords:** *Hevea brasiliensis*; Euphorbiaceae; Rubber latex; Lectin; Agglutinin; Rubber particle; Latex coagulation

Rapepun Wititsuwannakul, Kamonchanok Rukseree, Kamonwan Kanokwiroon, Dhirayos Wititsuwannakul, A rubber particle protein specific for Hevea latex lectin binding involved in latex coagulation,

**Phytochemistry**, Volume 69, Issue 5, March 2008, Pages 1111-1118, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2007.12.007.

(<http://www.sciencedirect.com/science/article/pii/S0031942207007236>)

**Abstract:**

In the first of this three paper series, an in vitro latex coagulation was shown to arise from aggregation of rubber particles (RP) and lutoid membranes. RP aggregation was shown to be induced by a specific Hevea latex lectin-like protein (HLL) present on the lutoid membrane. In this second paper, a binding protein (BP) ligand counterpart for HLL was identified. This RP-HLLBP, having a specific interaction, with HLL was isolated from RP and characterized. The protein was extracted from the small RP in the presence of a surfactant (0.2% Triton-X-100) and further purified to homogeneity. Purification steps included acetone precipitation, heat-treatment, and column chromatography. The presence of RP-HLLBP was monitored by its ability to compete with erythrocytes in the hemagglutination inhibition (HI) assay. The purified RP-HLLBP had an HI titre of 1.37 [ $\mu$ ]g ml<sup>-1</sup>, a pI value of 5.4, optimum activity at pH 5-8 and was thermostable up to 60 oC. On SDS-PAGE a single glycoprotein with Mr of 24 kDa was detected while on native PAGE the major Mr was about 120 kDa. The purified RP-HLLBP was shown to inhibit latex coagulation. Chitinase, but no other glycosidase tested, abolished its HI action and inhibited HLL-induced RP aggregation in a competitive dose dependent manner. This indicated the presence of, and role for, N-acetylglucosamine residues in the binding recognition. The Hevea latex lectin-like protein can thus be referred to as a Hevea latex lectin. Based on protein identification by peptide mass fingerprinting, the RP-HLLBP was confirmed to be the small rubber particle protein (SRPP). This work has unambiguously determined the role of an intrinsic RP glycoprotein (RP-HLLBP or SRPP) as a key component in formation of the rubber latex coagulum.

**Keywords:** *Hevea brasiliensis*; Euphorbiaceae; Rubber particle; Lectin receptor; Rubber particle protein; Rubber latex; Latex coagulation

K.M. Hosamani, K.S. Katagi, Characterization and structure elucidation of 12-hydroxyoctadec-cis-9-enoic acid in *Jatropha gossypifolia* and *Hevea brasiliensis* seed oils: a rich source of hydroxy fatty acid,

**Chemistry and Physics of Lipids**, Volume 152, Issue 1, March 2008, Pages 9-12, ISSN 0009-3084, DOI: 10.1016/j.chemphyslip.2007.11.003.

(<http://www.sciencedirect.com/science/article/pii/S0009308407004628>)

**Abstract:**

The seed oils containing hydroxy fatty acids are being used in various industries such as, in the production of nylon-11, in the manufacture of multi-purpose greases, as good anti-rust, new additives in water-soluble cutting fluids, and antimicrobial activity of various derivatives of ricinoleic acid polymers. *Jatropha gossypifolia* and *Hevea brasiliensis* seed oils were found to contain 18.5% and 18.0% of 12-hydroxyoctadec-cis-9-enoic acid (ricinoleic acid), respectively. The identification and characterization was based on UV, FTIR, <sup>1</sup>H NMR, <sup>13</sup>C NMR, MS, GC analysis and chemical degradations.

**Keywords:** *Jatropha gossypifolia*; *Hevea brasiliensis*; Seed oils; Fatty acids; Unusual fatty acid; 12-Hydroxyoctadec-cis-9-enoic acid; Industrial utilization

Wenshuang Xie, Colleen M. McMahan, Amanda J. DeGraw, Mark D. Distefano, Katrina Cornish, Maureen C. Whalen, David K. Shintani, Initiation of rubber biosynthesis: In vitro comparisons of benzophenone-modified diphosphate analogues in three rubber-producing species,

*Phytochemistry*, Volume 69, Issue 14, October 2008, Pages 2539-2545, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2008.07.011.

(<http://www.sciencedirect.com/science/article/pii/S0031942208003531>)

**Abstract:**

Natural rubber, cis-1,4-polyisoprene, is a vital industrial material synthesized by plants via a side branch of the isoprenoid pathway by the enzyme rubber transferase. While the specific structure of this enzyme is not yet defined, based on activity it is probably a cis-prenyl transferase. Photoactive functionalized substrate analogues have been successfully used to identify isoprenoid-utilizing enzymes such as cis- and trans-prenyltransferases, and initiator binding of an allylic pyrophosphate molecule in rubber transferase has similar features to these systems. In this paper, a series of benzophenone-modified initiator analogues were shown to successfully initiate rubber biosynthesis in vitro in enzymatically-active washed rubber particles from *Ficus elastica*, *Hevea brasiliensis* and *Parthenium argentatum*.

Rubber transferases from all three species initiated rubber biosynthesis most efficiently with farnesyl pyrophosphate. However, rubber transferase had a higher affinity for benzophenone geranyl pyrophosphate (Bz-GPP) and dimethylallyl pyrophosphate (Bz-DMAPP) analogues with ether-linkages than the corresponding GPP or DMAPP. In contrast, ester-linked Bz-DMAPP analogues were less efficient initiators than DMAPP. Thus, rubber biosynthesis depends on both the size and the structure of Bz-initiator molecules. Kinetic studies thereby inform selection of specific probes for covalent photolabeling of the initiator binding site of rubber transferase.

**Keywords:** *Parthenium argentatum*; *Guayule*; *Ficus elastica*; *Hevea brasiliensis*; Photoaffinity labeling; Benzophenone; Allylic pyrophosphate; Farnesyl pyrophosphate; Rubber transferase; Cis-prenyltransferase



Bernardo M.T. da Costa, Jay D. Keasling, Colleen M. McMahan, Katrina Cornish, Magnesium ion regulation of in vitro rubber biosynthesis by *Parthenium argentatum* Gray,

**Phytochemistry**, Volume 67, Issue 15, Rod Croteau Special Issue, Part 1, August 2006, Pages 1621-1628, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2006.04.010. (<http://www.sciencedirect.com/science/article/pii/S0031942206002160>)

**Abstract:**

Natural rubber is produced by a rubber transferase (a cis-prenyltransferase). Rubber transferase uses allylic pyrophosphate to initiate the rubber molecule and isopentenyl pyrophosphate (IPP) to form the polymer. Rubber biosynthesis also requires a divalent metal cation. Understanding how molecular weight is regulated is important because high molecular weight is required for high quality rubber. We characterized the in vitro effects of Mg<sup>2+</sup> on the biosynthetic rate of rubber produced by an alternative natural rubber crop, *Parthenium argentatum* (guayule). The affinity of the rubber transferase from *P. argentatum* for IPP [middle dot] Mg was shown to depend on the Mg<sup>2+</sup> concentration in a similar fashion to the *H. brasiliensis* rubber transferase, although to a less extreme degree. Also, in vitro Mg<sup>2+</sup> concentration significantly affects rubber molecular weight of both species, but molecular weight is less sensitive to Mg<sup>2+</sup> concentration in *P. argentatum* than in *H. brasiliensis*.

**Keywords:** *Parthenium argentatum*; *Compositae*; *Hevea brasiliensis* Muell. Arg.; *Euphorbiaceae*; Regulation; Rubber transferase; FPP; IPP; cis-Prenyltransferase; Magnesium

R. Lacote, O. Gabla, S. Obouayeba, J.M. Eschbach, F. Rivano, K. Dian, E. Gohet, Long-term effect of ethylene stimulation on the yield of rubber trees is linked to latex cell biochemistry,

**Field Crops Research**, Volume 115, Issue 1, 4 January 2010, Pages 94-98, ISSN 0378-4290, DOI: 10.1016/j.fcr.2009.10.007.

(<http://www.sciencedirect.com/science/article/pii/S0378429009002743>)

**Abstract:**

Ethylene stimulation with ethephon (2-chloroethylphosphonic acid) is nowadays essential for increasing latex production in the rubber tree (*Hevea brasiliensis*): both small-scale planters and agro-industrial plantations worldwide use ethephon. Ethylene stimulation strongly influences cumulative yield and latex cell biochemistry. The purpose of this study was to characterize the long-term behaviour of the rubber tree under ethephon treatment. Over a period of 7 years in Cote d'Ivoire, West Africa, the effect of eight frequencies of ethylene stimulation on yield and latex cell biochemistry was compared in four rubber trees clones, IRCA 130, IRCA 230, GT 1, and PB 217. The ability of the trees to produce more latex under ethylene stimulation was related to the sucrose and inorganic phosphorus contents of the latex cells. For high-yield clones with low sugar content and high inorganic phosphorus content like IRCA 130, namely quick starter clones, no stimulation was necessary to improve yield. For clones like IRCA 230, with higher sugar content, eight ethylene stimulations per year was the optimum frequency to obtain the highest yield. The effect of ethylene stimulation on latex yield increase was significant in clones with high sucrose content and low inorganic

phosphorus content such as PB 217. These clones, namely slow starter clones, needed more stimulation to produce more, but in the longer run there were no negative effects of ethylene on the latex yield. These results will help planters optimize latex production by choosing the most appropriate ethylene stimulation to clones according to their latex cells biochemistry.

**Keywords: Hevea brasiliensis; Ethylene stimulation; Latex cell biochemistry; Sucrose; Inorganic phosphorus; Latex yield**

## **PLANT PHYSIOLOGY-NUTRITION (6 jdl)**

Panida Kongsawadworakul, Unchera Viboonjun, Phayao Romruensukharom, Pisamai Chantuma, Somjintana Ruderman, Herve Chrestin, The leaf, inner bark and latex cyanide potential of *Hevea brasiliensis*: Evidence for involvement of cyanogenic glucosides in rubber yield,

***Phytochemistry***, Volume 70, Issue 6, April 2009, Pages 730-739, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2009.03.020.

(<http://www.sciencedirect.com/science/article/pii/S0031942209001344>)

### **Abstract:**

The latex of *Hevea brasiliensis*, expelled upon bark tapping, is the cytoplasm of anastomosed latex cells in the inner bark of the rubber tree. Latex regeneration between two tappings is one of the major limiting factors of rubber yield. *Hevea* species contain high amounts of cyanogenic glucosides from which cyanide is released when the plant is damaged providing an efficient defense mechanism against herbivores. In *H. brasiliensis*, the cyanogenic glucosides mainly consist of the monoglucoside linamarin (synthesized in the leaves), and its diglucoside transport-form, linustatin. Variations in leaf cyanide potential (CNp) were studied using various parameters. Results showed that the younger the leaf, the higher the CNp. Leaf CNp greatly decreased when leaves were directly exposed to sunlight. These results allowed us to determine the best leaf sampling conditions for the comparison of leaf CNp. Under these conditions, leaf CNp was found to vary from less than 25 mM to more than 60 mM. The rubber clones containing the highest leaf CNp were those with the highest yield potential. In mature virgin trees, the CNp of the trunk inner bark was shown to be proportional to leaf CNp and to decrease on tapping. However, the latex itself exhibited very low (if any) CNp, while harboring all the enzymes ([beta]-d-diglucosidase, linamarase and [beta]-cyanoalanine synthase) necessary to metabolize cyanogenic glucosides to generate non-cyanogenic compounds, such as asparagine. This suggests that in the rubber tree bark, cyanogenic glucosides may be a source of buffering nitrogen and glucose, thereby contributing to latex regeneration/production.

**Keywords: Hevea brasiliensis; Euphorbiaceae; Rubber tree; Latex; Leaf and bark cyanide potential; Cyanide metabolism; Linamarin; Rubber yield**

Rapepun Wititsuwannakul, Piyaporn Pasitkul, Kamonwan Kanokwiroon, Dhirayos Wititsuwannakul, A role for a Hevea latex lectin-like protein in mediating rubber particle aggregation and latex coagulation,

**Phytochemistry**, Volume 69, Issue 2, January 2008, Pages 339-347, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2007.08.019.

(<http://www.sciencedirect.com/science/article/pii/S0031942207005274>)

**Abstract:**

An in vitro aggregation of washed lutoid membrane and rubber particles, respectively, prepared from the bottom (lutoid) fraction and rubber layer of centrifuged fresh latex, leading to the formation of rubber coagulum necessary for a latex coagulation was demonstrated. A Triton X-100 extract of washed lutoid membrane proteins, isolated and prepared from the bottom fraction of centrifuged fresh latex was examined for its role in the latex coagulation process. It induced agglutination of rabbit erythrocytes, indicating the presence of a lectin-like protein. Hevea latex lectin-like protein (HLL) was purified to homogeneity by active chitin binding separation, followed by DEAE-Sepharose chromatography. Its Mr analyzed by SDS-PAGE was 17 kDa, whereas that determined by gel filtration was 267 kDa. The HLL had a pI value of 7.2. Several glycoproteins were shown to inhibit the HLL-induced hemagglutination. The hemagglutinin activity of HLL was enhanced by Ca<sup>2+</sup>. Of most interest was the finding that HLL strongly induced aggregation of the Hevea latex rubber particles (RP). This strong RP aggregation leads to latex coagulation, indicating the possibility that it is involved in the formation of the coagulum that plugs the latex vessel ends and stops the flow of latex upon tapping. In addition, the purified HLL also induced aggregation of RP taken from several other non-Hevea latex producing plants. This might indicate either a common or universal role of this lectin-like protein in RP aggregation and hence latex coagulation. This paper, for the first time, provides clear and unequivocal evidence for either a key biological role or physiological function of an endogeneous latex lectin-like protein in the sequential process of latex coagulation.

**Keywords:** *Hevea brasiliensis*; Euphorbiaceae; Rubber latex; Lectin; Agglutinin; Rubber particle; Latex coagulation

Rapepun Wititsuwannakul, Kamonchanok Rukseree, Kamonwan Kanokwiroon, Dhirayos Wititsuwannakul, A rubber particle protein specific for Hevea latex lectin binding involved in latex coagulation,

**Phytochemistry**, Volume 69, Issue 5, March 2008, Pages 1111-1118, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2007.12.007.

(<http://www.sciencedirect.com/science/article/pii/S0031942207007236>)

**Abstract:**

In the first of this three paper series, an in vitro latex coagulation was shown to arise from aggregation of rubber particles (RP) and lutoid membranes. RP aggregation was shown to be induced by a specific Hevea latex lectin-like protein (HLL) present on the lutoid membrane. In this second paper, a binding protein (BP) ligand counterpart for HLL was identified. This RP-HLLBP, having a specific interaction, with HLL was isolated from RP and characterized. The protein was extracted from the small RP in the presence of a surfactant (0.2% Triton-X-100) and further purified to homogeneity.

Purification steps included acetone precipitation, heat-treatment, and column chromatography. The presence of RP-HLLBP was monitored by its ability to compete with erythrocytes in the hemagglutination inhibition (HI) assay. The purified RP-HLLBP had an HI titre of 1.37 [ $\mu$ ]g ml<sup>-1</sup>, a pI value of 5.4, optimum activity at pH 5-8 and was thermostable up to 60 °C. On SDS-PAGE a single glycoprotein with Mr of 24 kDa was detected while on native PAGE the major Mr was about 120 kDa. The purified RP-HLLBP was shown to inhibit latex coagulation. Chitinase, but no other glycosidase tested, abolished its HI action and inhibited HLL-induced RP aggregation in a competitive dose dependent manner. This indicated the presence of, and role for, N-acetylglucosamine residues in the binding recognition. The Hevea latex lectin-like protein can thus be referred to as a Hevea latex lectin. Based on protein identification by peptide mass fingerprinting, the RP-HLLBP was confirmed to be the small rubber particle protein (SRPP). This work has unambiguously determined the role of an intrinsic RP glycoprotein (RP-HLLBP or SRPP) as a key component in formation of the rubber latex coagulum.

**Keywords: Hevea brasiliensis; Euphorbiaceae; Rubber particle; Lectin receptor; Rubber particle protein; Rubber latex; Latex coagulation**

Zhi-Li Zhang, Jia-Hong Zhu, Quan-Qi Zhang, Yuan-Bao Cai, Molecular characterization of an ethephon-induced Hsp70 involved in high and low-temperature responses in *Hevea brasiliensis*,

*Plant Physiology and Biochemistry*, Volume 47, Issue 10, October 2009, Pages 954-959, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2009.06.003.

(<http://www.sciencedirect.com/science/article/pii/S0981942809001454>)

**Abstract:**

Hsp70s have been shown to play important roles in helping cells to cope with adverse environments, especially in response to temperature. In this study a novel ethephon-induced Hsp gene, designated as HbHsp70, was isolated from *Hevea brasiliensis*. The HbHsp70 cDNA contained a 1965 bp open reading frame encoding 655 amino acids. The deduced HbHsp70 protein showed high identities to Hsp70s from other plants. Expression studies revealed more significant accumulation of HbHsp70 transcripts in leaves and stems than in roots, barks and latex. The transcription of HbHsp70 was induced by ethephon, heat treatment and low temperature stress, whereas jasmonic acid had little effects. Recombinant HbHsp70 was expressed in *Escherichia coli* and purified by Ni-NTA affinity chromatography. Measuring the light scattering of luciferase (Luc) revealed that HbHsp70 prevents the aggregation of luc during high-temperature stress. In vitro experiments showed that HbHsp70 had protective functions not only against heat stress but also against chilling stress. All these data suggest that HbHsp70 may play roles in responses to heat shock and low temperature in *H. brasiliensis*.

**Keywords: Hevea brasiliensis; Heat shock protein 70; Ethephon; Expression; Low temperature**

Zhi-Li Zhang, Jia-Hong Zhu, Quan-Qi Zhang, Yuan-Bao Cai, Molecular characterization of an ethephon-induced Hsp70 involved in high and low-temperature responses in *Hevea brasiliensis*,

***Plant Physiology and Biochemistry***, Volume 47, Issue 10, October 2009, Pages 954-959, ISSN 0981-9428, DOI: 10.1016/j.plaphy.2009.06.003.

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**Keywords:** *Hevea brasiliensis*; Heat shock protein 70; Ethephon; Expression; Low temperature

Rapepun Wititsuwannakul, Piyaporn Pasitkul, Pattavuth Jewtragoon, Dhirayos Wititsuwannakul, Hevea latex lectin binding protein in C-serum as an anti-latex coagulating factor and its role in a proposed new model for latex coagulation, ***Phytochemistry***, Volume 69, Issue 3, February 2008, Pages 656-662, ISSN 0031-9422, DOI: 10.1016/j.phytochem.2007.09.021.

(<http://www.sciencedirect.com/science/article/pii/S0031942207005936>)

**Abstract:**

A distinct protein specifically recognized by its strong interaction with Hevea latex lectin (HLL) was detected in the aqueous C-serum fraction of centrifuged fresh latex. This C-serum lectin binding protein (CS-HLLBP) exhibited strong inhibition of HLL-induced hemagglutination. The CS-HLLBP was purified to homogeneity by a protocol that included ammonium sulfate fractionation, size exclusion and ion exchange chromatography. The purified CS-HLLBP had a specific HI titer of 0.23 [ $\mu$ ]g ml<sup>-1</sup>. Its Mr analyzed by SDS-PAGE was ca. 40 kDa and that by gel filtration was ca. 204 kDa. It has a pI value of 4.7, an optimum activity between pH 6 and 10 and was heat stable up to 50 [degree sign]C. The HI activity of CS-HLLBP was abolished upon treatment with chitinase. The CS-HLLBP inhibited HLL-induced rubber particle aggregation in a dose dependent manner. A highly positive correlation between CS-HLLBP activity and rubber yield per tapping was found. The correlations for fresh latex ( $r = 0.98$ ,  $P < 0.01$ ) and dry rubber ( $r = 0.95$ ,  $P < 0.01$ ) were both highly significant. This indicated that the CS-HLLBP might be used as a reliable marker for the mass screening of young

seedlings to identify and select clones with potential to be superior producers of rubber. A latex anti-coagulating role of the CS-HLLBP is proposed. The findings described in this 3 paper series have been used to propose a new model of rubber latex coagulation that logically describes roles for the newly characterized latex lectin and the two lectin binding proteins.

**Keywords:** *Hevea brasiliensis*; Euphorbiaceae; Rubber latex; C-serum; Lectin; Lectin binding protein; [alpha]-Globulin; Latex flow; Anti-coagulating factor; Latex coagulation

## **PLANT PHYSIOLOGY-GROWTH AND DEVELOPMENT (2 jdl)**

Terry Coffelt, Lauren Johnson, A set of descriptors for evaluating guayule germplasm, *Industrial Crops and Products*, In Press, Corrected Proof, Available online 14 May 2010, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2010.04.004.

(<http://www.sciencedirect.com/science/article/pii/S092666901000083X>)

### **Abstract:**

Commercialization of guayule (*Parthenium argentatum* Gray) as a source of rubber is receiving world-wide attention as an alternative to *Hevea* in order to meet increasing demand for natural rubber. As more breeders, agronomists, botanists and other scientists become involved in evaluating and developing guayule germplasm, it is imperative that a uniform set of germplasm descriptors is available. These descriptors are also necessary for use in obtaining plant variety protection certificates. The objective of the current study was to develop a set of descriptors for evaluating guayule germplasm in breeding and agronomic studies as well as identify a minimum set of descriptors to use when time or labor make using all of the descriptors not feasible. Based on the experience of the authors and previous research, a set of descriptors was developed and used to evaluate breeding nurseries of both traditionally and transgenically developed guayule plants. The descriptors were easy to use and required a minimum amount of time per plant, so that a large number of lines could easily be evaluated. The descriptors adequately covered the range of diversity observed in the nurseries evaluated. A set of 14 minimum descriptors is proposed - plant height, plant width, biomass, number of main branches, bark thickness, leaf shape, leaf serration number, dormancy break, flower bud score, 1000 seed weight, rubber content and yield, and resin content and yield. For germplasm protection uses, the full set of descriptors should be used. The descriptors will provide uniformity in comparing germplasm performance across environments. The descriptors will also be valuable to regulatory agencies in granting various plant variety protection certificates.

**Keywords:** *Breeding*; *Germplasm evaluation*; *Hypoallergenic latex*; *Plant growth*; *Biomass*

P. Dissanayake, D.L. George, M.L. Gupta, Effect of light, gibberellic acid and abscisic acid on germination of guayule (*Parthenium argentatum* Gray) seed, *Industrial Crops and Products*, Volume 32, Issue 2, September 2010, Pages 111-117, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2010.03.012. (<http://www.sciencedirect.com/science/article/pii/S0926669010000798>)

**Abstract:**

Guayule (*Parthenium argentatum* Gray) produces high quality, low-allergenic rubber which has commercial potential. The species has small seeds with a high level of dormancy which was investigated in a series of germination experiments. Many researchers have investigated the effect of gibberellic acid and light on guayule seed dormancy but there have been no reports on light quality. In this study, increased germination was found for yellow (82.0%) and red (65.3%) light compared to green (55.3%) and blue (25.3%) light. Effect of light quality indicates evidence of phytochrome-mediated germination and dormancy in guayule. A higher ratio of red to far-red radiation in yellow and red light activates phytochrome that stimulates production of endogenous gibberellins to promote germination. The highest level of far-red radiation found in blue light had similar inhibitory effects as complete darkness (21.3%). An intermediate level of red to far-red radiation for green light produced intermediate germination. Seed coat and light also affected germination of freshly harvested guayule seed. Embryos responded to light compared with darkness (68.0% vs. 34.7%) albeit to a much lower level than intact seed (57.3% vs. 8.6%) indicating the possible presence of inhibitors in the seed coat. Light and GA3 strongly interact to overcome dormancy in guayule and appear to act on the same pathway. The optimum concentration of GA3 for seed used in this experiment ranged from 250 to 500 ppm.

**Keywords: Guayule; Dormancy; Light quality; Gibberellic acid; Abscisic acid; Seed coat**

## PLANT PHYSIOLOGY-REPRODUCTION ( 2 jdl)

D. Jasso de Rodriguez, J.L. Angulo-Sanchez, R. Rodriguez-Garcia, An overview of guayule research and development in Mexico, *Industrial Crops and Products*, Volume 24, Issue 3, 2005 Annual Meeting of the Association for the Advancement of Industrial Crops: The International Conference on Industrial Crops and Rural Development, November 2006, Pages 269-273, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2006.06.018. (<http://www.sciencedirect.com/science/article/pii/S0926669006000926>)

**Abstract:**

Shrubs grown at Navidad, Nuevo Leon, were identified with up to 21% rubber content. This may be the result of plants been subject to high levels of soil water stress, or it may be genetically inherited. The rubber quality was also affected by environmental factors, as shown by multimodal molecular weight curves from determined by gel permeation chromatography. Plants from the Mapimi site were identified as diploids which are sexually reproducing, compared to other ploidy levels that reproduce by apomixis, a

form of clonal propagation. This is significant because diploids population are not known to occur in other regions of the world, and are useful for plant breeding. Irrigation treatments significantly influenced rubber and resin yield by increasing the plant biomass. However, rubber and resin content were not affected.

**Keywords:** Guayule; Native collections; Rubber; Resin; New crop

Cesar A. Reyes-Lopez, Martha Pedraza-Escalona, Guillermo Mendoza, Alejandra Hernandez-Santoyo, Adela Rodriguez-Romero, A single amino acid substitution on the surface of a natural hevein isoform (Hev b 6.0202), confers different IgE recognition, *FEBS Letters*, Volume 580, Issue 10, 1 May 2006, Pages 2483-2487, ISSN 0014-5793, DOI: 10.1016/j.febslet.2006.03.085.

(<http://www.sciencedirect.com/science/article/pii/S0014579306004121>)

**Abstract:**

Decreased immune reactivity of isoforms of major allergens has been reported. However, such claims have always been based on experiments with recombinant proteins. This work describes the molecular and physicochemical characterization of a hevein (Hev b 6.0201) natural isoform (Hev b 6.0202), which is present in rubber latex from *Hevea brasiliensis*. The isoallergen has a single substitution Asn14Asp, which gives rise to local differences in the surface potential, as observed from the crystal structure presented here. Besides, ELISA inhibition using serum pools of adult and pediatric patients showed reduced IgE-binding capacity (~27%) with the isoallergen. Overall, these results are relevant to delineate crucial residues involved in this dominant discontinuous epitope.

**Keywords:** Crystal structure; Isoallergens; Hevein; Hev b 6.0201; IgE-epitope

## PEST OF PLANTS (1 jdl)

Nestor Laurier Engone Obiang, Georges Salle, Faut-il eradiquer *Phragmanthera capitata*, parasite des heveas en Afrique ?,

*Comptes Rendus Biologies*, Volume 329, Issue 3, March 2006, Pages 185-195, ISSN 1631-0691, DOI: 10.1016/j.crv.2006.01.007.

(<http://www.sciencedirect.com/science/article/pii/S1631069106000229>)

**Abstract:**

The effect of *Phragmanthera capitata* (Sprengel) Balle, an epiphytic Loranthaceae, on latex production of three clones of rubber tree, *Hevea brasiliensis* (Mull. Arg.), was studied in the industrial plantation of Mitzic, in Gabon. The 22-year-old trees, presenting the largest trunk circumference, were the most parasitized and at the same time the best producers of latex. At first, this would suggest that mistletoe parasitism could have a favourable effect on latex yield. However, an accurate statistical analysis showed that, in fact, parasitism had no significant effect on latex yield. These data, discussed in relation to the biology of flowering parasitic plants, suggest that there is no urgency to eradicate *Phragmanthera capitata* for the moment. To cite this article: N.L. Engone Obiang, G. Salle, C. R. Biologies 329 (2006).

**Keywords:** Loranthaceae; Production; Hevea; Angiosperme parasite; Gabon; Loranthaceae; Production; Rubber tree; Parasitic plant; Gabon



## PLANT DISEASES (1 jdl)

Vincent Le Guen, Jean Guyot, Carlos Raimundo Reis Mattos, Marc Seguin, Dominique Garcia, Long lasting rubber tree resistance to *Microcyclus ulei* characterized by reduced conidial emission and absence of teleomorph,

***Crop Protection***, Volume 27, Issue 12, December 2008, Pages 1498-1503, ISSN 0261-2194, DOI: 10.1016/j.cropro.2008.07.012.

(<http://www.sciencedirect.com/science/article/pii/S0261219408001282>)

### **Abstract:**

The resistance of the MDF 180 *Hevea brasiliensis* cultivar to South American leaf blight caused by *Microcyclus ulei* has been studied under both controlled conditions of inoculation, and under natural infestation in the Brazilian state of Bahia. Inoculated isolates on this cultivar in most cases developed sporulating lesions and produced conidiospores, but at a moderate level, characteristic of a race non-specific, partial resistance. The teleomorph of *M. ulei* has never been reported on this cultivar, either in controlled conditions or in natural infestation, whereas it is commonly observed on other susceptible cultivars. This type of resistance has seldom been described for other pathosystems. MDF 180 resistance having also endured for more than 30 years in areas very conducive to the disease can, therefore, be described as a durable resistance. Rubber productivity of this cultivar is not sufficient to allow planting on industrial scale, but its resistance characteristics make it a good progenitor candidate for rubber tree resistance breeding programs.

**Keywords:** *Hevea brasiliensis*; South American leaf blight; Partial resistance; Durable resistance

## SOIL CHEMISTRY (1jdl)

Akbar Najafi, Ahmad Solgi, Seyed Hamidreza Sadeghi, Soil disturbance following four wheel rubber skidder logging on the steep trail in the north mountainous forest of Iran, ***Soil and Tillage Research***, Volume 103, Issue 1, April 2009, Pages 165-169, ISSN 0167-1987, DOI: 10.1016/j.still.2008.10.003.

(<http://www.sciencedirect.com/science/article/pii/S0167198708001906>)

### **Abstract:**

Reduced soil porosity, water content, forest floor mass and rutting are four common disturbances during timber harvesting and skidding. The present study was conducted to evaluate effects of skid trail slope and traffic frequency on soil disturbance in a temperate mountainous forest. Four levels of traffic (3, 7, 14 and 20 passes of a rubber skidder HSM 904), and three levels of slopes (gentle <10%, moderate 10-20%, steep >20%) were applied in three replicates consequently, 36 plots with 10 m long by 4 m wide were utilized in the study. All samples were taken from four randomized 4 m-transects in each plot across the wheel track perpendicular to the direction of travel with 2 m buffer zone between lines to avoid interactions. Results showed that during skidding total porosity, soil water and Forest floor decreased significantly ( $P \leq 0.05$ ). Average total porosity on the treatments of >20% slope reduced significantly ( $P \leq 0.05$ ) in comparison with those of <20% slope through all level of traffics. The lowest total

porosity was identified in treatment with slope >20% and 14 passes by soil compaction. Soil water content was decreased up to 45% after skidding in comparison with undisturbed area. There was a negative correlation between water content and skid trail slope. The forest floor mass decreased on steep trail during skidding in all traffics. The amount of forest floor mass on the treatments under the 7 passes and slopes of >20% treatments (437.6 kg ha<sup>-1</sup>) was significantly ( $P \leq 0.05$ ) lower than those under the 14 passes and slopes of <10% (841.4 kg ha<sup>-1</sup>). Rut depth were recorded in 14 passes and increased with slope (18 cm < 10%, 27.5 cm in 10-20% and 34.5 cm in >20%). In 3 passes, increase of skid trail slope from 10% to 20% corresponded to a decrease of 15%, 22%, and 67% in total porosity, water content and forest floor mass, respectively. The results indicated that slope steepness had a strong effect on the soil physical properties. In the present study, the skidder extracted different volume of logs during skidding operation. In order to obtain better interpretation the skidder must carry a fixed load during operation.

**Keywords: Litter mass; Soil compaction; Skid trail slope; Soil disturbance; Soil wetness; Iranian forest**

## **SOIL FERTILITY (1jdl)**

Hua Zhang, Gan-Lin Zhang, Yu-Guo Zhao, Wen-Jun Zhao, Zhi-Ping Qi, Chemical degradation of a Ferralsol (Oxisol) under intensive rubber (*Hevea brasiliensis*) farming in tropical China,

***Soil and Tillage Research***, Volume 93, Issue 1, March 2007, Pages 109-116, ISSN 0167-1987, DOI: 10.1016/j.still.2006.03.013.

(<http://www.sciencedirect.com/science/article/pii/S0167198706000717>)

### **Abstract:**

Impacts of intensive management practices on rubber (*Hevea brasiliensis*) farms (e.g., land clearance, fertilization, and rubber tapping) have not been adequately investigated. In this study, soil was taken from fields of grass (before rubber plantation), immature rubber (before tapping), and mature rubber (after tapping) at a tropical farm in Hainan, China. Soil organic matter, plant nutrients, cations, and soil pH were determined. Rubber cultivation resulted in significant decline of soil organic C and microbial biomass C. Available P was extremely low for all soils, resulting from the naturally low P content and the high sorption capacity of highly weathered Ferralsol. Furthermore, soil pH decreased by about 0.5 units, accompanied by an increase of exchangeable Al by more than one-fold. Regression analysis demonstrated that soil acidification was characterized by the depletion of base cations and release of Al. To maintain tropical soil quality, farming practices such as liming and organic amendment should be included in the best management practices of rubber farm.

**Keywords: Ferralsols; Oxisols; Hevea brasiliensis; Sustainable management; Soil quality; Soil acidification; Tropics**

## AGRICULTURAL PRODUCT (3 jdl)

Robert G. Hamilton, Katrina Cornish, Immunogenicity studies of guayule and guayule latex in occupationally exposed workers,

***Industrial Crops and Products***, Volume 31, Issue 1, January 2010, Pages 197-201, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2009.09.012.

(<http://www.sciencedirect.com/science/article/pii/S0926669009001812>)

### **Abstract:**

Type I *Hevea brasiliensis* rubber latex allergy is managed by avoidance, using synthetic and alternative latex (such as *Parthenium argentatum*, guayule) products. This study investigates the ability of high-dose occupational exposure to guayule shrub/homogenate/latex to induce guayule-specific antibody responses in employees (Yulex Corporation). Participants completed an allergy history/guayule exposure questionnaire and provided annual blood samples from 2006 to 2008. Sera were analyzed for IgG and IgE anti-guayule (protein from homogenate, commercial-grade latex and non-ammoniated total plant proteins) using solid phase immunoassays (negative = IgG < 1 [ $\mu$ g/ml, IgE < 1 ng/ml). Guayule-specific IgG antibody (range: 2.0-9.7 [ $\mu$ g/ml) was detected in 3 of 16 (19%) highly exposed employees in the pilot plant and R&D/applications laboratory. Antibody levels related to relative cumulative-years (e.g. >3) of reported guayule homogenate/latex exposure. Equivocal IgG antibody responses (1.0-2.0 [ $\mu$ g/ml) were detected in 2 of 5 (40%) of administrators with infrequent guayule homogenate/latex contact. No guayule-specific IgE antibody or guayule-associated allergic reactions were detected. We conclude that protein from guayule and in guayule latex can be immunogenic but not allergenic in occupationally exposed workers.

**Keywords:** *Hevea brasiliensis*; *Parthenium argentatum*; Guayule; Latex allergy; IgE antibody; IgG antibody; Occupational exposure

C.R. Benedict, Rachel Goss, M.A. Foster, Paul J. Greer, The formation of rubber particles in developing cortical parenchyma of *Parthenium argentatum* plants exposed to the low temperatures of fall and winter of the Chihuahuan Desert,

***Industrial Crops and Products***, Volume 30, Issue 3, November 2009, Pages 403-406, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2009.07.009.

(<http://www.sciencedirect.com/science/article/pii/S0926669009001216>)

### **Abstract:**

In this paper we have examined rubber particle formation in cortical parenchyma cells of guayule (*Parthenium argentatum* Gray) plants exposed to the low temperature of the fall and winter of the Chihuahuan Desert that stimulates rubber formation.

Plants were transplanted to field plots in May in Ft. Stockton, TX. In September most of the cortical parenchyma in cross-sections 3 cm from the stem tips and from the base of the stems were mature, containing a central vacuole, tonoplast and a parietal cytosol with chloroplasts, mitochondria, peroxisomes and a few rubber particles. In 8-month-old plants examined in January many of the cortical parenchyma cells 3 cm from the stem tip were immature and completely filled with cytosol containing a high population of

rubber particles. At this stage of development there was a thickened secondary cell wall but no central vacuole, tonoplast or parietal cytosol and all the rubber particles were formed in the central cytosol. In the cortical parenchyma in early stages of central vacuole development, by the coalescing of microvacuoles, the rubber particles were located in the developing central vacuole and in the cytosol being appressed toward the cell wall. In 8-month-old plants in January, the cortical parenchyma at the base of the stems was more mature than cells near the stem tips. These cells showed a later stage of development of a central vacuole from microvacuole fusing and contained a high population of mixed sizes of rubber particles and a thin layer of parietal cytosol containing chloroplasts, mitochondria and rubber particles with a partly developed tonoplast. There was clear evidence of the fusion of small rubber particles with other rubber particles that may represent a mechanism for the growth of rubber particles.

**Keywords: Guayule; Parthenium argentatum; Cortical parenchyma;  
Rubber particle ontogeny**

T.A. Coffelt, F.S. Nakayama, D.T. Ray, K. Cornish, C.M. McMahan, Post-harvest storage effects on guayule latex, rubber, and resin contents and yields, *Industrial Crops and Products*, Volume 29, Issues 2-3, March 2009, Pages 326-335, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2008.06.003.

(<http://www.sciencedirect.com/science/article/pii/S092666900800126X>)

**Abstract:**

Guayule is a new crop being commercialized for hypoallergenic latex production. Because natural processes that occur in the plant following harvest, notably dehydration, result in rapid loss of latex and immediate processing of guayule shrub for latex on a commercial scale is not feasible, storage conditions that maintain latex concentration and yield need to be established. The objective of this study was to determine the effects of different storage conditions on the extractable latex, total rubber, resin, and guayulin A and B contents, and extractable latex, total rubber, and resin yields in harvested guayule shrub. The experiment was established using plants transplanted into the field at the University of Arizona Maricopa Agricultural Center, Maricopa, AZ, USA, on 22 March 2001. A randomized complete block design with four replications was used. Two germplasm lines (11591 and AZ-2) were used for this experiment. Twenty plants of each line were harvested six times (November 2002, March 2003, July 2003, November 2003, March 2004, and July 2004) from each field plot. Two plants of each line were randomly assigned to each of 10 storage treatment combinations reflecting wet, dry, or wet alternated with dry conditions prior to chipping for latex extraction. Extractable latex content, total rubber content, resin content, and guayulin A and B contents were determined after storage and compared with freshly harvested shrub. Plant biomass, latex yield, rubber yield, and resin yield were also determined and compared with fresh harvested shrub. AZ-2 was significantly lower in latex, rubber, and guayulin A content than 11591, and significantly higher in biomass, latex yield, rubber yield, resin content, resin yield, and guayulin B content. The results from this study show that moist storage of harvested shrub prior to dry chipping allows a higher yield of latex. Storing the shrub under moist conditions may allow more flexible

harvesting and processing schedules, by limiting post-harvest latex losses and increasing the time interval between harvesting and processing.

**Keywords:** *Parthenium argentatum*; Shrub storage; Guayule; Rubber yield; Latex yield; Rubber content; Latex content; Resin yield; Resin content; Guayulin content

## STORAGE AGRICULTURAL PRODUCTS (1 jdl)

T.A. Coffelt, F.S. Nakayama, D.T. Ray, K. Cornish, C.M. McMahan, Post-harvest storage effects on guayule latex, rubber, and resin contents and yields, *Industrial Crops and Products*, Volume 29, Issues 2-3, March 2009, Pages 326-335, ISSN 0926-6690, DOI: 10.1016/j.indcrop.2008.06.003.

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### **Abstract:**

Guayule is a new crop being commercialized for hypoallergenic latex production. Because natural processes that occur in the plant following harvest, notably dehydration, result in rapid loss of latex and immediate processing of guayule shrub for latex on a commercial scale is not feasible, storage conditions that maintain latex concentration and yield need to be established. The objective of this study was to determine the effects of different storage conditions on the extractable latex, total rubber, resin, and guayulin A and B contents, and extractable latex, total rubber, and resin yields in harvested guayule shrub. The experiment was established using plants transplanted into the field at the University of Arizona Maricopa Agricultural Center, Maricopa, AZ, USA, on 22 March 2001. A randomized complete block design with four replications was used. Two germplasm lines (11591 and AZ-2) were used for this experiment. Twenty plants of each line were harvested six times (November 2002, March 2003, July 2003, November 2003, March 2004, and July 2004) from each field plot. Two plants of each line were randomly assigned to each of 10 storage treatment combinations reflecting wet, dry, or wet alternated with dry conditions prior to chipping for latex extraction. Extractable latex content, total rubber content, resin content, and guayulin A and B contents were determined after storage and compared with freshly harvested shrub. Plant biomass, latex yield, rubber yield, and resin yield were also determined and compared with fresh harvested shrub. AZ-2 was significantly lower in latex, rubber, and guayulin A content than 11591, and significantly higher in biomass, latex yield, rubber yield, resin content, resin yield, and guayulin B content. The results from this study show that moist storage of harvested shrub prior to dry chipping allows a higher yield of latex. Storing the shrub under moist conditions may allow more flexible harvesting and processing schedules, by limiting post-harvest latex losses and increasing the time interval between harvesting and processing.

**Keywords:** *Parthenium argentatum*; Shrub storage; Guayule; Rubber yield; Latex yield; Rubber content; Latex content; Resin yield; Resin content; Guayulin content