DEPARTMENT OF AGRICULTURE
SOUTH AUSTRALIA

Plant Industry Division Report

THE PROGRESS TOWARDS COMMERCIALIZATION OF THE GUAYULE PLANT (*Parthenium argentatum*) AS A SOURCE OF RUBBER IN THE UNITED STATES OF AMERICA.

by

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Guayule
Parthenium argentatum)

A ten month old guayule plant (approximately 75 cm high) growing under irrigation at Bakersfield, California, U.S.A.

A close-up of guayule flower heads.

Guayule variety trials at the Schafter Research Station, Bakersfield, California.

The centre row of native guayule is suffering from various diseases on relatively heavy soils and under irrigation compared with the surrounding hybrid with *Parthenium stramonium*.

McFarland Research Station, Bakersfield, California.

THE PROGRESS TOWARDS COMMERCIALIZATION OF THE GUAYULE PLANT *(Parthenium argentatum)* AS A SOURCE OF RUBBER IN THE UNITED STATES OF AMERICA.

IMPLICATIONS FOR SOUTH AUSTRALIA

A.F. Tideman.

1. INTRODUCTION:

At the first meeting of the South Australian Technical Committee on Guayule Rubber Production held on the 22nd April, 1981 (see Appendix I) it was decided that approval should be sought for me to extend a proposed private visit to the United States of America to assess the current research programmes associated with the commercialization of guayule with a view to advising that Committee whether renewed attempts, abandoned since World War II, should be undertaken to produce rubber from guayule in South Australia.

In particular it was agreed that I should endeavour to:-

1. Alert the American Joint Commission on Guayule Research and Commercialization that South Australia is again interested in developing this industry after carrying out detailed research during the 1940's.

2. Study the breeding methodology being used to improve latex production.

3. Examine agronomic aspects of the field establishment and general husbandry of the crop.

4. If possible gain access to breeding material and seed supplies.

Subsequently, Cabinet approval was given for me to discuss developments in Washington with the Manager of the National Science Foundation and with the Executive Secretary and Programme Manager on the Joint Commission on Guayule Research and Commercialization, (Dr. Richard Wheaton). Approval was also given to study the research programmes in progress at four centres in California and Arizona.

My itinerary, including the personnel with whom I worked, is recorded in Appendix II.

The following Report covers the technical information I gained from my visits and discussions. It concentrates on the agronomic aspects of guayule development. I have analysed this information as far as possible to assess its relevance to South Australia. I have also included complete lists of references so that this report can be used as the basis for a research and development programme if and when it becomes a reality in South Australia without the Report unnecessarily repeating details already documented.

At the time of writing the South Australian Technical Committee is awaiting further information before advising the Government whether to go ahead and if so, what resources will be needed. The information will be gained from a study of the bio-climatic requirements of guayule in relation to the Australian environment soon to be completed by Dr. Henderson, CSIRO, who has been working in the U.S.A. in recent months and from a planned visit by the American Joint Commission on Guayule planned for late March, 1982. Also the South Australian Technical Committee has made arrangements for Mr. Ragless, its Executive Officer, to visit New South Wales in March, 1982 to assess field developments there which have been in progress since September, 1980.

2. THE WORLD RUBBER SITUATION - GUAYULE POTENTIAL:

Natural rubber is vital to Australia. We are totally dependent on foreign sources of supplies. America, in the same position, has recognised this and taken positive steps through the Native Latex Act of 1978 to commercialize guayule. Congress has approved expenditure totalling $5M (US) this financial year for research.

Guayule potential, in relation to the world rubber supplies, has been previously reviewed by Mr. Ownes in a report submitted at the first meeting of the South Australian Technical Committee. It is an excellent report which makes it unnecessary for me to comment further except to say that all the details I gathered while in America only substantiate what he has already reported. Three members of the Joint Commission on Guayule Research and Commercialization reviewed this report with me and fully substantiated its contents. Twenty-five copies were made available to the Commission.

In brief, there is considerable evidence that an 'OPEC' rubber situation could develop for Australia by 1990.

REFERENCES

I have selected the following references to give a broad introduction to the potential for the commercialization of guayule.

General Publications

1. American National Research Council (Board on Science and Technology for International Development) "Guayule: An Alternative Source of Natural Rubber". 1977. Library of Congress Catalogue No. 76 -62525.

2. Cornforth, C.C.; R.D. Lacewell and G.S. Collins "Guayule - Economic Implications of Production in the Southwestern United States". Texas Agricultural Experiment Station, MP-1466, 1980.

3. Foster, K.E. *(et al.)*. "A Technology Assessment of Guayule Rubber Commercialization". Office of Arid Lands Studies, University of Arizona, Tucson, Arizona, and Midwest Research Institute, Kansas City, Missouri. May 1980.

4. Foster, K.E. *(et al.)*. "The Role of Guayule in Meeting Future U.S. Rubber Demand". Elastomerics, April 1981.

5. Hammond, B.L. and L.G. Polhamus. Research on Guayule *(Parthenium argentatum)* 1942-1959 Technical Bulletin No. 1327, Agricultural Research Service, U.S. Department of Agriculture, Washington, D.C. 1965.

6. Polhamus, L.G. (1962). "Rubber Botany Production Utilization".

7. Native Latex Commercialization and Economic Development Act of 1978. Public Law 95-592, U.S. Congress.

8. Stewart, G.A. *(et al.)*. The Potential for Production of Hydrocarbon Fuels from Crops in Australia Report. Department of National Development and Energy, Australia Government Publishing Service, 1982.

9. El Guayulero. A Publication of the Guayule Rubber Society. Available from the Los Angeles Arboretum, 301N Baldwin Avenue, Arcadia, CA 91006.

10. Williams, G. *(et al.)*. "Guayule Rubber Production - The World War II Emergency Rubber Project - A Guide to Future Development". Office of Arid Land Studies, University of Arizona, Tucson, Arizona, U.S.A.

11. Jensen, M.E. (1981). "Review of Guayule Research". USDA Science and Education Administration.

12. Weih and Nivert, (April 1980). "Assessing Guayule's Potential. An Up-date of Firestone's Development Programme and Economic Studies".

South Australian Publications

13. Crocker, R.L. and H.P.C. Trumble (1945). "Investigations of Guayule in South Australia". CSIR Bulletin 192.

South Australian Publications contd.

14. Ellis, M.R. "Guayule". A report to the Assistant Director, Economic Studies 6.1.78. Department of Economic Development, South Australian Government Docket No. DED 138/77.

15. Owens, L.W. (1981). "Report on Guayule Potential and Research in South Australia". Mines & Energy Department.

16. Prescott, J.A. (1943). "Australian Homoclimes of the Zone of Natural Occurrence of *Parthenium argentatum*". Transactions of the Royal Society of South Australia 67 (2) 312.

17. Shapter, R.E. (1952). "Rubber Accumulation in Guayule in South Australia". CSIRO Bulletin 270.

18. Australian Plant Production Committee. Reports from States to the Committee, September, 1977. Sixth meeting, Agenda Item 11.

3. THE GUAYULE PLANT:

3.1 Description and Classification

Guayule is a bushy perennial shrub with alternate narrow leaves along the stem, and covered in a drought-protecting white wax. It bears a canopy of small flowers borne on exceptionally long stems. It is inconspicuous, hardy and between two and three feet high. It can survive 30 - 40 years under desert conditions.

Guayule flowers are pollinated by wind and by insects. Their tiny seeds are produced at a prolific rate. After a single rainfall, a single plant can yield several thousand seeds. Vigorously growing plants bloom and set seed continuously throughout summer and spring. If the seeds are stored carefully, they can remain viable for several decades. Flowers and seeds are produced as early as six months after germination.

Guayule*(Parthenium argentatum)* is so named because of a silvery sheen on its gray-green leaves. The genus Parthenium is a member of the sunflower family, Compositae. There are sixteen species of Parthenium.

Guayule is closely related to parthenium weed *(Parthenium hysterophorus)* which has been proclaimed a primary pest plant on Schedule I of the Pest Plants Act (S.A.) 1975. This is a serious weedy species first found in Australia in Queensland in 1955 and is now widespread. It is a serious weed in Asia and North America. There is however no evidence that guayule will become weedy.

3.2 Latex Characteristics of Guayule

Of the sixteen species of Parthenium, guayule is the only species known to produce rubber in any quantity.

Unlike the rubber in Hevea and other latex-producing plants, guayule rubber is not contained in ducts, but in single, thin-walled cells. These rubber-filled cells are mainly in the outer layers (in the cortical tissues and the medullary rays) and mostly in new-grown tissues, but the old cells of the inner xylem and pith produce rubber for several years. Two-thirds of the rubber is in the stems and branches; the remainder in the roots. There is no rubber in the leaves. Resin ducts are found throughout the shrub, and resins constitute 10-15 per cent of the plant dry weight.

In the native guayule bushes, rubber constitutes about 10 per cent of the total weight of the plant (dry weight). The strains that were widely cultivates in America in the 1940's were reported to be able to produce 20 per cent rubber (dry weight) after four years growth.

Rubber is not metabolized or used by the plant because it continues to accumulate for at least 10 years, and plants can remain in the field for even longer periods without serious loss of rubber.

When guayule is growing actively it produces little or no rubber.

3.3 Distribution

Guayule grows wild in some semi-arid regions of North America such as the Stockton Plateau and Big Bend region of Texas. It is native to the upland plateaus in Mexico and Texas with suptropical- temperate climates, where rainfall is low and erratic. Here it grows in a wide variety of shallow, stony, calcareous, and friable soils.

The areas where it can be grown under irrigation in the United States of America is Shown on Map !.

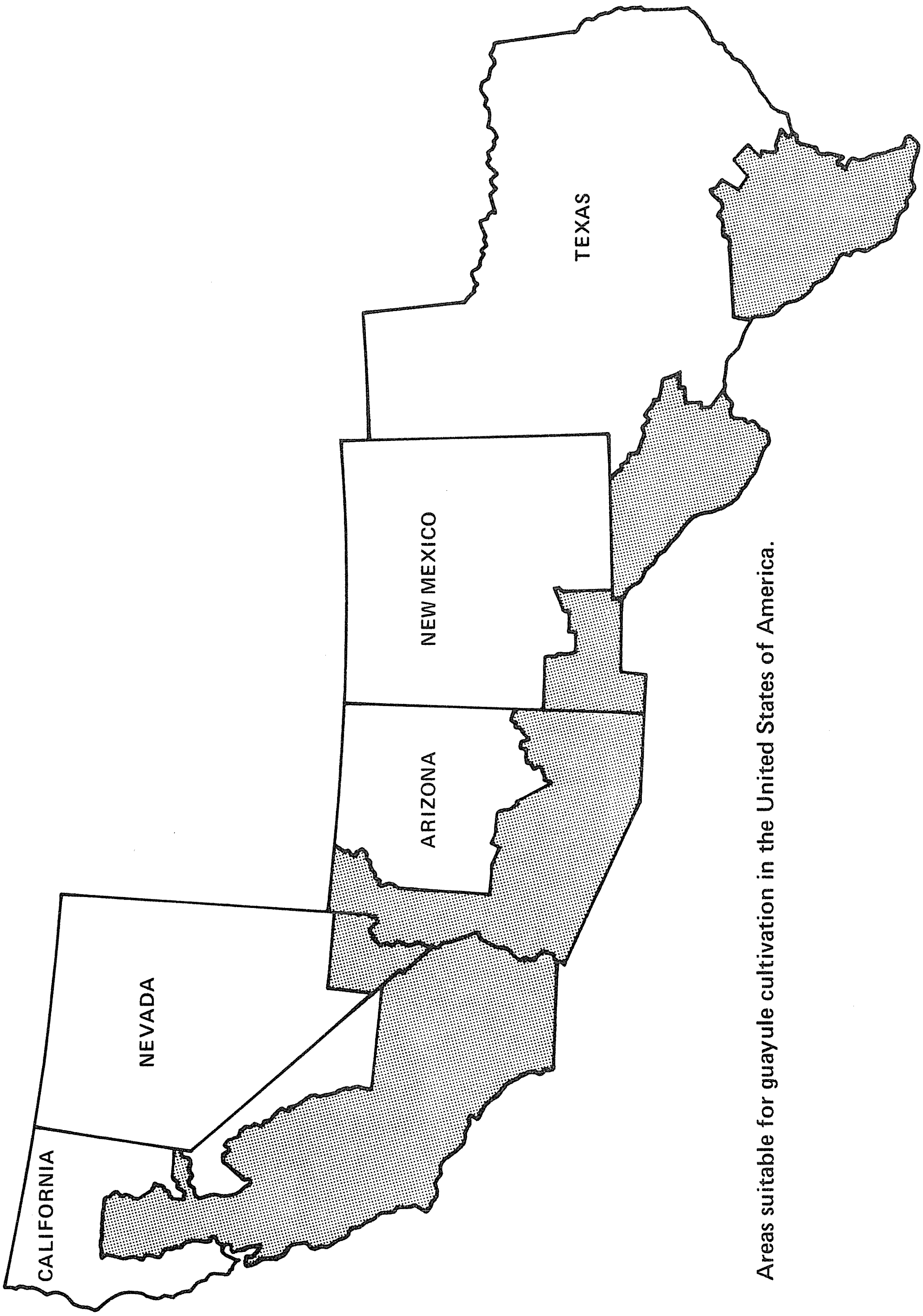
3.4 The Genetics of Guayule

Guayule can be readily manipulated by plant breeders for improvement because there exists many strains of wide genetic diversity. Diversity ranges from 2n = 36 to plants with chromosome numbers of 100 or more. Strains carrying 36 chromosomes are completely sexual. Plants of higher chromosome numbers are apomicts. Diploid guayule is largely self-sterile. Because of this the back-crosses with guayule and sibling crosses of hybrids show some degree of incompatability.

4. THE ADMINISTRATION OF THE AMERICAL GUAYULE RESEARCH AND DEVELOPMENT PROGRAMME.

The administration, research and development of guayule in America prior to the introduction of the Native Latex Commercialization Act of 1978 is largely summarized in one volume, USDA Technical Bulletin 1327 (See Reference 5 below) and needs no further elaboration.

In March, 1979, the Joint Commission on Guayule Research and Commercialization was established under the terms of the Native Latex Commercialization Act of 1978. It consisted of three members from the United States Department of Agriculture, three from the Department of Commerce, one from the National Science Foundation and one from the Bureau of Indian Affairs.



Areas suitable for guayule cultivation in the United States of America.

A new chairman, Dr. A.R. Bertrand, Director of Science and Education was appointed Chairman of the Commission, now known as the Joint Commission on Native Latex in January, 1981. The members of the Joint Commission and its staff group are listed in Appendix III and Appendix IV respectively.

The Commission is currently supporting the following nine projects at an approximate cost of $1.5 M (U.S.) annually.

Project Title	Objectives	Supervisor
Identification and control of guyaule diseases.	(a) To identify the pathogens which currently are causing diseases of native and experimental commercial stands of guayule in the south-western United States and in Mexico. (c) To evaluate the effects of diverse soil-inhabitants of bacteria on the growth responses of guayule and on pathogens of guayule. (c) To also attempt to effect control of pathogens through chemical treatments and/or development of resistant lines. Chemical controls will be particularly orient-ed towards damping off organisms, search for resistance will initially focus on controlling.	Dr. Stanley Marcus ALCORN, Plant Pathology Dept., University of Arizona, Tucson, Arizona. 85721
Water Use & Production Practices for Guayule.	The proposed research is intended to investigate modern production aspects of guayule culture. Research conducive to the establishment, growth and harvest of guayule will be conducted. Investigators in agronomy, and irrigation will work cooperatively to evaluate the agronomic practices necessary if guayule is to become a commercial crop of arid and semi-arid regions.	Dr. Delma D.FANGMEIER, Soils, Water & Engineering Dept., College of Agric. University of Arizona, Tucson, Arizona. 85721
Seed Production Practices for Guayule Commerciali-zation.	(a) Determine field management practices for seed yield and seed quality. (b) Develop procedures for harvesting threshing and cleaning seed and determine the effectiveness and efficiency of various machinery and equipment in the cmmercialization of each operation. (c) Determine optimum specification for treatment of seed dormancy, for seed germination tests and for seed storage conditions, and determ-ine what characteristis determine seed quality.	Dr. David D. RUBIS, Plant Sciences Dept. College of Agric.(AES) University of Arizona, Tucson, Arizona. 85721

Project Title	Objectives	Supervisor
Guayule Improvement Utilizing the Arizona Germplasm Collections	1. Evaluate experimental lines and germplasm for optimum rubber yield. 2. Improve water use efficiency and drought tolerance. 3. Improve seed and seedling vigor. 4. Increase disease resistance in seedling and mature plants. 5. Broaden the spectrum of genetic variability through intra and inter-specific hybridization. 6. Herbicide and salt tolerance evaluation.	Duane Lee JOHNSON Plant Sciences Dept. College of Agric.(AES) University of Arizona, Tucson, Arizona. 85721
Breeding for Increas -ing Rubber Yield in Guayule	Development of higher rubber yielding varieties using inter and intraspecific hybridization.	Dr. George P. HANSON, Los Angeles State & County Arboretum, 301N. Baldwin Ave. Arcadia, California. 91006
Optimizing Rubber Production in Guayule by Breeding & Selection.	Development of higher rubber yielding varieties using inter and intraspecific hybridization.	Hewitt M. TYSDAL, Div. of Plant Industry California Dept. of Food & Agriculture, 5960 Encina Road, Goleta, California. 93017
Developing Salinity Irrigation & Fertil -ity Management of Guayule Grown with Gypseous Saline Water.	1. Evaluate salt tolerance of guayule, including salt effects on rubber yields. 2. Evaluate water requirement and water stress effects on rubber yields under the influence of salinity. 3. Develop and test appropriate irrigation schedules for growing guayule with gypseous saline waters. 4. Evaluate nitrogen and phos- phorus requirement for growing guayule.	Seiichi MIYAMOTO, Soils & Water Engineer ing Dept. El Paso Expt. Station, Texas A & M Univ. El Paso, Texas.

Project Title	Objectives	Supervisor
Establishment and Cultivation Practices for Guayule Production in West Texas.	1. Estimation of the effect of planting date, irrigation methods, and seed treatment on field establishment by direct seeding. 2. Estimation of the effect of irrigation level and plant spacing on rubber and seed production. 3. Survey and selection of apparent high-yielding individuals from native populations in Texas and increasing seed stock of these and prior selections. 4. Collection and identification of insects present in field guayule and evaluation of real and potential damage from each species.	Dr. Jimmy L. TIPTON, Research Center at El Paso, Texas Agric. Expt. Station, Texas A & M Univ. El Paso, Texas. 79927.
Influence of Establishment Methods & Herbicides on Guayule Rubber Production.	1. Determine the most effective method of direct field seeding of guayule. 2. Develop more effective herbicide combinations for weed control in direct seeded and transplant guayule. 3. Compare rubber production of directed seeded vs transplant guayule grown under weed control regimes appropriate to each method of culture.	Dr. J. Wayne WHITWORTH, Dept. of Agronomy, New Mexico Agric. Exp. Station, New Mexico State Univ. Las Cruces, New Mexico. 88003

Since 1979 at a further total cost of approximately $1.75M (U.S.) the National Science Foundation has supported the following projects.

Investigator	Institution	Grant Period	Purpose
K. Foster	U. Arizona	9/78-11/79	Technology Assessment of Guayule Commercialization.
K. Foster	U. Arizona[b]	8/80-9/81	Technology Assessment for Mexico.
D. Bragg	TX & A & M.U.	1/79-3/80	Analysis of Market and Production cost of Guayule Rubber.
D. McIntyre	U. Akron[b]	5/76-4/78 5/78-4/80 8/80-8/82	Physical Properties and Efficacy of Guayule Rubber Efficacy of Guayule and By Products.

b. Joint Project with Centro de Investigacion en Quimica Applicada (CIQA), Saltillo, Mexico.

Investigator	Institution	Grant Period	Purpose
P. Phillips	U. Utah[b]	7/80-8/82	Guayule rubber Crystallization
J. White	U. Tenn.[b]	6/80-6/82	Rheological Properties of Guayule Rubber .
A. Verbiscar	Anver Bio.	1/80-1/81	Guayule By-products and Rubber Analysis .
D. Rubis	U. Arizona	9/76-2/78 9/79-9/80	Seed Collection and Study Genetics, Cytogenetics and Breeding
G. Hanson	L.A. Arboretum	4/77-3/78 4/78-3/79 4/79-3/80 4/80-11/80	Seed Collection, Breeding, Selection and Horticultural Studies.
H. Yokoyama G. Hanson	L.A. Arboretum USDA/SEA	5/78-4/79 5/79-5/80 5/80-12/80	Bioregulator Treatment for Enhancement of Rubber Yield.
T. Murashige	U. Calif. Riverside	12/78-11/79 1/80-1/81	Regeneration of Guayule Plants from Cell and Tissue Culture.
E. Radriguez	U. Calif. Irvine	2/79-1/80	Ascertain Allergencitiy of Guayule.
H. Bloss	U. Arizona	3/79-9/80	Mycorrhizal Associations in Guayule and Jojoba.
C. Benedict	TX A & M U.	8/79-8/80	Regulation of Rubber Formation in Guayule.
D. Fangmeier	U. Arizona	9/79-9/80	Water Use and Production Practices.
W. Whitworth	N.M. State U.	9/79-9/80	Herbicides for Weed Control in Guayule Plantations.

Finally, the Department of Commerce commencing in February, 1981, have allocated $200 000 to the Institute of Polymer Science at the Akron University, Ohio to study uses for the resins and non-rubber components of the plant and the harvesting-processing interface.

The American Administration has a written agreement with Mexico for joint research and development of guayule. A further agreement is being negotiated with the New South Wales Government.

5. FIRESTONE'S DEVELOPMENT PROGRAMME IN THE UNITED STATES OF AMERICA:

In November, 1977, the Firestone management approved a project to explore the commercial potential for guayule. The project consists of an agricultural programme located at the Firestone Test Centre at Fort Stockton, Texas, and a process and product development programme being carried out at their laboratories in Akron. The Company plans to have a pilot plant for rubber extraction operating at Akron during 1982.

The current position of the Company is summarised in their 1980 report as follows:-

"At present, economic indicators appear quite favourable; however cautious optimism should be exercised in light of the heavy initial capital investment, the risky long-term pay back and the unforeseen complications which will arise.

Based on experience gained, and the favorable outlook, Firestone is committed to guayule".

While in the United States I did not have the opportunity to examine private development projects at first hand, however my discussions lead me to interpret the above statement to mean that the Company will be looking for solid Government backing before it is committed to guayule commercialization.

6. AMERICAN RESEARCH PROJECTS:

6.1 University of California - Davis - Dr. Knowles

Unfortunately, because of the Mediterranean fruit-fly outbreak near San Francisco, I was not able to work with Dr. Siddigni who controls the guayule programme for the Californian Department of Agriculture and Food in the vicinity of Sacramento and more northern areas of California. However I was able to study the programme at the Davis campus of the University of California where Dr. Paul Knowles is comparing four selections of guayule with a bulk native seed source for rubber production under various fertilizer and irrigation applications. He is also comparing the effectiveness of various herbicides.

I was only able to study in detail the herbicide programme. I selected this knowing that during the war in South Australia weed control was an over-riding serious problem which was not satisfactorily answered and I am sure this would have prevented successful commercialization if that had finally been attempted.

Guayule is slow growing and is quickly overgrown and shaded by weeds. If shaded guayule dies and only a very low density of weeds can severely stunt the plants.

At Davis mechanical weed control has been compared with 13 herbicides. It has been proved that there is no possible economic way that weeds can be mechanically controlled in guayule plantations. However the trials have isolated five of the 13 herbicides treated as having practical use with no damage to the newly planted guayule.

The pre-plant herbicide trifluralin has proved very useful although of course it must be incorporated into the soil. Simazine, oryzalin, nitrofen and oxyfluorfen have proved successful as post-planting herbicides which can be sprayed over the guayule. The weeds relevant to South Australia for which they were achieving good control using these chemicals included pigweed *(Amaranthus* spp.), crab grass *(Digitaria sanguinalis)*goosefoots *(Chenopodium* spp.) field bud weed *(Convolvulus arvensis)* and a wide range of grasses such as brome grass and ryegrass.

The trials have shown that dalapon bromoxynil glyphosphate and 2,4- D severely damaged guayule on rates necessary for world control.

6.2 Los Angeles State and County Arboretum - Dr. G.P. Hanson

At the Los Angeles State and County Arboretum a team of five senior research officers and seven well qualified technical staff, lead by Dr. George Hanson, are concentrating on Guayule research using a gene pool established in 1978 following detailed field surveys in Mexico.

The team has excellent facilities using the grounds of what we would call the botanic gardens in which to plant out their field trials. One would think these would be too public for safety but they do not seem to have any problems.

Their work has been in progress since 1975 and now covers a wide range of investigations which I tried to absorb in the day visit. Their breeding programme is the most significant.

6.2.1 Breeding Programme

The team is investigating the underlying mechanisms of inheritance in guayule. They have identified genetic markers including isozymes, trichome, morphology and phytochemical characteristics en-a b ling segregation to be analysed.

From the very variable material available from the desert, selection programmes for high ribber, vigor, cold tolerance, disease resistance and resistance to moving have indexed plants that have been used in back crossing and for hybridization with *Parthenium incanum, P. fruitcosuiim* and *P . tomentosum*.

I believe these programmes will inevitably produce cultivars with much higher rubber production but three to five years more work will be needed before results come to hand.

6.2.2 Physiological Studies and Agronomic Studies

The team has been concentrating on the development of seed collection, cleaning and germination procedures, all of which are now well understood and sound technology has been developed.

Their treatment of guayule seed for dormancy is worth recording in this text:- (Harvesting and cleaning techniques are covered in the references.

6.2.3 Treating Guayule Seed for Seed Dormancy

Procedure:

1. Soak seed in water for 8 hours and wash.

2. Soak in NaOCl for 2 hours.

   Concentration depends on age of seeds:

(a)	Freshly harvested seeds	1.0% NaOCl
(b)	6 month old seeds	0.5% NaOCl
(c)	One year or older seeds	0.25% NaOCl

3. Wash in water and dry.

The team has also carried out studies of guayule growth under greenhouse conditions to develop alternate propagation systems (cutting, pollarding) including tissue culture which requires special comment. During the week of my visit Dr. Dave Radin, who is responsible for this work, reported that he had been able to produce tissues on culture carrying considerable rubber content an unusual achievement as secondary metabolic products are not often developed on cultures. This of course raises the interesting question of producing rubber in 'plant factories'.

6.2.4 Disease Studies

Guayule is seriously attacked by two diseases, Verticillium wilt and Phytophthora root rot disease. This team at Los Angeles is studying these fungi and has isolated various strains, *Parthenium icanum* and *P. confertium* two species closely related to guayule are highly resistant to both diseases.

6.2.5 Rubber Analysis

Rubber analysis is a very important aspect of any breeding project and I attempted with members of this team and with help from Dr. Tysdal.

This team at Los Angeles follow an analysis procedure based on Traub's photometric analysis. This is probably the best being relatively cheap - quick.

The other methods available are the C_{13} - NMR analysis, the soxhlet extraction method and the soxhlet extraction method and the blender analysis.

Using the analysis technique described in Appendix V the team seems to be making good progress in selecting germplasm of high rubber yielding plants.

6.3 USDA - Cotton Research Station - Shafter - Dr. Tysdal

On 16th July, 1981, I was able to visit the USDA Cotton Research Station at Shafter which is on the outskirts of Bakersfield in California, to discuss the guayule research and development programme with Dr. Tysdal.

Dr. Tysdal has a life time experience in guayule research which commenced at the beginning of World War II. It was he who selected, in the early 1050's the best 25 cultivars of guayule then known and ensured that this germ plasm was put 'on-ice' for future needs. In this he showed a great deal of foresight because the mood of the USDA at that time was to have nothing more to do with the plant.

Dr. Tysdal, although retired, has an assistant, Dr. Ali Estilai, and together they have an extensive breeding programme at Shafter and at the McFarland outstation 20 miles to the north-east of Bakersfield.

The overall concept of the guayule research and development programme is outlined diagramatically in Appendix VI.

At the McFarland outstation, these workers are concentrating on the diploids of *Parthenium argentatum,* I noted in these lines excellent and wide variation from which they are selecting lines for disease resistance and high rubber yield. Some lines are yielding, in 75 cm rows with plants 45 cm apart irrigated at 10 day intervals, 10-15% rubber. The site is particularly prone to verticillium wilt and phytophthora which provides obvious screening with excellent resistance evident in some lines.

At the same site these duploid lines are being crossed with *P. stramonium.*

The hybrids *P. argentatum x P. stramonium, P. argentatum x P. fruticosum* and *P. argentatum x P. mariola* are also being studied.

At Shafter, on the outskirts of Bakersfield, where the U.S. Cotton Research Station is established I was able to visit a variety trial site using 11 of the 25 lines selected by Dr. Tysdal after World War II, designated N 396 and N 565 are giving highest rubber yields under irrigation.

At the Centre, Dr. Tysdal is developing the use of Nuclear Magnetic Resonance to give early rubber tests on live plant tissue without extraction. I believe there are grave doubts about the reliability of this technique.

Currently Dr. Tysdal is using a simple extraction process for determining the rubber content of his lines by chopping 20 grams of stem from secondary developed branches to 1.5 cm pieces. These are vitamized with a 50% water and 50% ethyl alcohol solution for 14 minutes. Salt is added and the rubber plus resin coagulates as a green lump on the surface. This is filtered off, dried and weighed.

I had the opportunity of studying a seed thrashing machine and a seed harvester developed at the Centre (See Appendix VII).

The use of coagulators to stimulate yield and quality of rubber is also being examined.

6.4 University of Arizona

Dr. David Rubis with Dr. Ray and Miss Massey, assisted me over a two day period, to gain an insight into the research programme for guayule development at the University of Arizona. I visited the trial sites at Yuma and Mesa.

Basically they are involved in developing techniques for seed build-up and also the breeding of improved cultivars particularly using hybridization.

Another unit at the University is involved in the identification and control of guayule diseases.

The group lead by Dr. Rubis has successfully adapted a Bud Antel brocholli planter to plant guayule seedlings in the field.

I believe one of the most important contributions this team is currently making to the development of guayule as a source of rubber are the trials associated with harvesting a different level above he roots and studying the various cultivars capacities to regenerate. It will apparently be possible to mow fields as we do for lucerne allowing rubber to be harvested without the destruction of the stand with the subsequent need for replanting. These trials are still inconclusive.

Reference

The Proceedings of the Third International Guayule Conference held at Pasadena California between 27th - 30th April, 1980 contains a complete and up-to-date series of scientific papers covering breeding methods, including hybridization, weed and disease control, seed technology and harvesting techniques. It gives an excellent basis for selected reading for any research and development programme planned for South Australia. I am holding a full set of reprints.

APPENDIX I

SOUTH AUSTRALIAN ADVISORY COMMITTEE ON GUAYULE COMMERCIALIZATION

Technical Secretary: SAGRIC	Mr. David C. Ragless, Department of Agriculture, G.P.O. Box 1671, ADELAIDE. 5001
Members:	Mr. A.F. Tideman, Chief, Plant Industry Division, Department of Agriculture, ADELAIDE. 5001
W.A.R.I.	Dr. J. Silsbury, Senior Lecturer, Waite Agricultural Research Institute, GLEN OSMOND. 5064
CSIRO Div. of Hort.	Dr. J.S. Hawker, Sen. Principal Research Off. Scientist, CSIRO Division of Horticulture, GLEN OSMOND. S.A. 5064
S.A. Inst. of Technology.	Dr. R.E. Underdown, Principal Lecturer, Chemical Technology, The Levels Campus, P.O. Box 1, POORAKA. S.A. 5095
	Dr. Ron Watts, Lecturer in Chemical Technology, The Levels Campus, P.O. Box 1, POORAKA. S.A. 5095
Roseworthy Agricultural College	Dr. V.R. Squires, Dean of Faculty (Natural Resources), Roseworthy Agricultural College, ROSEWORTHY. S.A. 5371
Department of Mines & Energy	Mr. L. Owens, Manager, Energy Development, Department of Mines & Energy, 191, Greenhill Road, PARKSIDE, S.A. 5063
Department of Trade & Industry	Ms Palmer, Project Officer, Department of Trade & Industry, G.P.O. Box 1264, ADELAIDE. 5001
State Development Office	Mr. N. Lawson, Chief Project Officer, State Development Office, G.P.O. Box 1264, ADELAIDE. 5001
Bridgestone Aust. Pty. Ltd.	Mr. R.A. Footner, Managing Director, Bridgestone Aust. Pty. Ltd. 1028,South Road, EDWARDSTOWN. 5039

Date	Location	Contact	Programme
26/6/81	Washington	Dr. R. Wheaton, Programme Manager, Domestic Rubber Programme, USDA.	Administrative overview of technical programmes.
		Dr. D. McClintock, Food & Agricultural Adv. Department of State.	Co-operative research between the U.S.A. and South Australia
		Dr. R. Neetz, Deputy Director-Scientific Exchange, USDA.	As above
		Dr. H. Hill, National Oceanic & Atmosphere Administration, USDA.	World production locations for guayule
		Dr. R. Brock, Scientific Counsellor Australian Embassy.	US-Australia agreements for co-operative research . Possible exchange programmes.
29/6/81	Washington	Dr. H. Huang, Programme Manager, National Science Foundation.	Guayule Research overview. US funding.
10/7/81	Davis, California	Dr. P. Knowles, Professor of Agronomy, Agronomy Department, University of California.	Guayule cultivar comparative trials. Fertilizer rates. Irrigation techniques. Weed control.
11/7/81 (A.M.ONLY)	As above	As above	As above
12/7/81 13/7/81 14/7/81	Returned to San Francisco	Trade Commission Officer, Industry inputs to guayule. Mr. D. Fagg.	Dr. Siddigni, Californian Department of Food & Agriculture planned to show me the research and development work in northern California but was unavailable to do so due to the Mediterranean Fruit Fly outbreak.
15/7/81	San Francisco Travelled to Los Angeles	Trade Commission Officer, Mr. Abrahams.	Review of industry inputs to the guyaule programme.
16/7/81	Bakersfield	Dr. Tysdal Dr. Estilai	Guayule breeding programme.

Date	Location	Contact	Programme
17/7/81	Los Angeles Arboretum	Dr. Hanson	Germ Plasm bank
		Dr. Radin	Hybridization
		Dr. Nagri	Cytogenetic studies
		Dr. Mehta	Pathological studies
		Dr. Banigan	Rubber analysis
		Dr. Verbiscau	Rubber analysis
18/7/81 19/7/81	Tuscon Arizona	Weekend	
20/7/81	Tuscon University	Dr. D. Rubis Dr. D. Day	Seed increase programme. Plant breeding
21/7/81	San Francisco	Report to the Trade Commission Office Mr. D. Fagg.	
23/7/81 24/7/81 25/7/81 26/7/81			Travelled to Australia. Schedule delayed by fuel strike.

APPENDIX III

MEMBERS OF JOINT COMMISSION ON NATIVE LATEX COMMERCIALIZATION

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APPENDIX IV

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APPENDIX V

Guayule Rubber Extraction and Analysis - The Technique Used at the Los Angeles Arboretum

Rubber in guayule is found most abundantly within the parenchyma cells of the stems and roots. It is locked-up there in such a way that the cell walls must be broken to get the rubber out. This is unlike the conventional "rubber tree", *Hevea braziliensis* where rubber flows with the latex through canals and can be tapped so that the latex flows out when the canals are severed. To break the cells open in guayule, stems are milled or ground to almost a powder. The leaves contain little or no rubber and are discarded soon after harvesting.

Rubber can be extracted from milled guayule in two ways. The first is a simple method that was used extensively in the rubber mills of Mexico and Texas in the early 1900's. The guayule shrub was processed in a pebble mill in the presence of water. The pebbles acted to comminute the shrub. During the process rubber gathered together in small particles while the woody and fibrous material was ground finer and finer. After a specified time the comminuted shrub was discharged into a tank of water. Rubber particles floated to the surface and the other material sank. The rubber was skimmed from the surface and pressed into blocks for shipment to the market.

The second extraction method requires the use of organic solvents to remove rubber from milled guayule. The plant tissue is first bathed in acetone to remove resinous materials, waxes, an pigments which are undesirable by-products for rubber production. Acetone is a convenient solvent to remove these compounds because rubber is not soluble in it; and it is relatively safe and inexpensive. The guayule material is next bathed in a suitable rubber solvent such as dichloromethane, tetrahydrofuran, benzene derivatives, or others which are highly volatile and hence can be evaporated off leaving the pure rubber. The solvent vapors may be condensed and reused many times.

Appropriate laboratory rubber analysis procedures can be derived from either of these two basic extraction methods. For example, the blendor extraction is a modification of the old pebble mill-floatation method described above. Here par-boiled stems are ground in a 50% alcohol + 50% water solution. The blendor breaks open the cells and allows the rubber to float out. The alcohol dissolves the undesirable compounds.

Solvent extraction lends itself readily to a variety of analysis procedures. The one described below has the advantages of being rapid, and accurate and are able to run up to fifty samples at a time. Here the solvent is relatively non-volatile - we don't want it to evaporate. To the rubber solution we add a precipitating agent which causes the rubber to come out of solution and remain as a colloidal suspension for a time. The turbidity of the suspension (turbidity is proportional to rubber concentration) is measured on a spectrophotometer from which rubber concentration may be calculated.

The following rubber analysis procedure is modified from: Traub, H.P. 1946. Rapid Photometric Methods for Determining Rubber and Resins in Guayule Tissue and Rubber in Crude-Rubber Products. U.S. Department of Agriculture Technical Bulletin 920. Modifications were developed by Dr. George P. Hanson, Mrs. Surinder Dhillon, and Miss Colette Beaupre.

I. PREPARATION OF DRIED SAMPLE

a. Branches 0.5 - 1.0 cm diameter are cut finely by clippers.
b. Pieces are dried in a 65°C forced air-drying oven for 24 hours to several days until grindable.
c. Dried samples are ground in an intermediate Wiley Mill using a No. 10 mesh screen.
d. Ground dried samples are again dried in the oven to give a constant weight.

II. USE OF THE WILLEMS POLYTRON

a. To a clean 50 ml centrifuge tube are added:

   1. 1.0 gm dried sample
   2. 10.0 ml diisobutyl ketone

b. The contents are ground using a Brinkman-Willems Polytron (PT 10-35 basic assembly, and PT 20 ST generator) for 15 seconds at a setting of 7.
c. The polytron blades are rinsed with about 10 ml of diisobutyl ketone into the above extract.

III. CENTRIFUGATION

a. Diisobutyl ketone extract is centrifuged at 1370G for 16 minutes.
b. The supernatant is transferred to a culture tube.
c. The residue is reground in 10 ml diisobutyl ketone using the polytron. The polytron blades are rinsed with diisobutyl 1370 G for 16 minutes. The supernatant is then added to the supernatant of the first extraction.
d. The solution is filtered through Whatman No. 1 filter paper into a 50 ml volumetric flask. The volume is made up to 50 ml with diisobutyl ketone.

NOTE: At this point, if necessary, the extract can be stored overnight in the refrigerator. No change in readings was observed in previous trials.

IV. PHOTOMETRY

a. 2.0, 0.5, or 0.2 ml extract is pipetted into a photometric tube. To this is added:

   1. 0 ml diisobutyl ketone to 2.0 ml extract, or 1.5 ml diisobutyl ketone to 0.5 ml extract, or 1.8 ml disobutyl ketone to 0.2 ml extract, and
   2. 2 drops colloidal solution
   3. 6.0 ml acidified alcohol

b. The solution is hand agitated for 30 seconds.
c. After 20 minutes, %-transmittance readings are taken on a B & L Spectronic 20 colorimeter.
d. Readings are read on a standard curve (made up using purified guayule rubber) and converted to rubber per cent on a dry weight basis.

Dilution factors for determining rubber concentration:
- 2.0 ml extract = x1
- 0.5 ml extract = x4
- 0.2 ml extract = x10

V. REAGENTS FOR RUBBER ANALYSIS

diisobutyl ketone = 2,6-dimethyl-4-heptanone, $(CH_3)_2CHCH_2\ COCH_2\ CH\ (CH_3)_2$ (note that diisobutyl ketone expands and contracts appreciably with temperature change).

colloidal solution = stereate solution = protective colloidal solution $O\ (CH_2CH_2OCC_{17}H_{35})\ 2$

made by dissolving 4.5 gm diglycerol stereate in 100 ml absolute alcohol by heating, and then making a 5% solution of this by volume in the ketone. (note that the colloidal solution may precipitate out at lower temperatures, and require warming to bring back into solution).

acidified alcohol = precipitant solution
0.5% solution of H_2SO_4 by volume in 95% ethanol (i.e. 5 ml: 995 ml)

Dennis Perry and George P. Hanson,
Los Angeles State and County Arboretum
Arcadia, California. 91006.

APPENDIX VI

THE TYSDAL GUYAULE DEVELOPMENT PROGRAMME BAKERSFIELD, CALIFORNIA

1979	1980			1981(*)	1982		1983		1984	1985		1986
Isolation Plots Arboretum	Jan-June	July	October	All year	April	July-Oct.	January	April	All year	April	July-Oct	and later
F1 P.stramonium x guayule interplanted with diploid guayule Planted in '79.	Produce seed	Greenhouse planting of 50000 seedlings for 5 acres	Transplant to 5 acre field F2s and BX1s	Rogue 99% by visual rubber disease. Treat with Bioregulator in Sept.	Transplant 500,(1%) selected F2s and BX1s to isolation plot with selected diploid guayule	Produce seed	Greenhouse planting for 50000 seedlings for 5 acres	Transplant to 5 acre field F3s and BX2s	Rogue 99% visual rubber disease Treat with Bio-regulator in Sept.	Transplant 500 F3 and BX2s	Produce seed Keep individual plants	Test all progeny of individ-ual plants for common variety. Many should be apomictic
F1 P.fruticosum x guayule interplanted with diploid guayle planted.	Same procedures as above. Selection in advanced generations and backcrosses.			Seed available for 5 acres in 1980	Make control crosses of exceptional F2 or BX1 pls with superior 54 or 72 chr. guayule for possible commercial variety.							
P.incanum x guaylle (not planted)	Cold resistant, 36 chromosome mariola should be crossed with selected diploid guayule and carried forward the same as above. There is no such plot at the Arboretum now.											
Diploid guayule Planted in '79.	Eliminate all but very best 36, (sexual) chr. guayule by intensive selection programme. These are the building blocks for improved guayule, higher rubber, etc. These plants are to be used in the interspecific crosses. Never in the history of guayule breeding has there been a consistent effort in this direction.											
Selected strains planted '79	Best strains from the Salinas breeding programme 1949-57 selected for this composite. Eleven strains were selected and there should be some selection among these for rubber and vigor.								Increase seed for New Variety			
Polyploid Guayule Planted '79.	Select best strains or plants in this group, but less effort here because selecting in apomictic strains is large a waste of time.											

(*) It would be better to have a second full year for selection but in a "Crash" programme it might be possible to speed up programme.

GUAYULE SEED HARVESTER

The Tysdal Guayule Seed Harvester has four essential elements:

(1) HOOD: approximately 26 inches by 26 inches at the base, built cone-shaped to come up to about 8 x 8 inches, or rounded, at the top. It has a "bonnet" in front with rolled side extenders to pick up side branches. It is built of fairly light metal, with good bracing and hanging support. It should be adjustable as to height, and sideways adjustment for different row widths.

(2) SUCTION FAN: should have plenty of volume, more than blowers (below) to provide a constant flow of air throughout the hood towards the top. Preferably by-pass, such as a cotton picker.

(3) BLOWERS: This is an unique feature, and important. Experience has shown that blowing is more effective than suction in large spaces. The blowers enter at the bottom of the hood at both sides to direct the seed upward, where the suction takes over. The suction should be in greater volume than the blowing. With foliage interference the blower is also more effective. Blowers were used effectively in the machine I built at Salinas in the 50s. One blower may suffice.

(4) VIBRATOR: This is important. The Denholm flower seed suction harvester which Keefer recently demonstrated on guayule at Lost Hills proved that the "vibrator" is effective in dislodging ripe seed. The vibrator has a sideways movement of about 3 or 4 inches at about 120 moves per minute. This however should be adjustable so that it can be speeded up when dislodgement is tougher, such as sometimes happens in the fall. It shakes the plant, but does not harm it. The vibrator does not break the branches or harm the tender peduncles of green flowers. The reel-type can cause damage and should not be used.

Other considerations

Clearance must be adequate so as not to damage plants, going over them several times a year. 40 inches would seem sufficient.

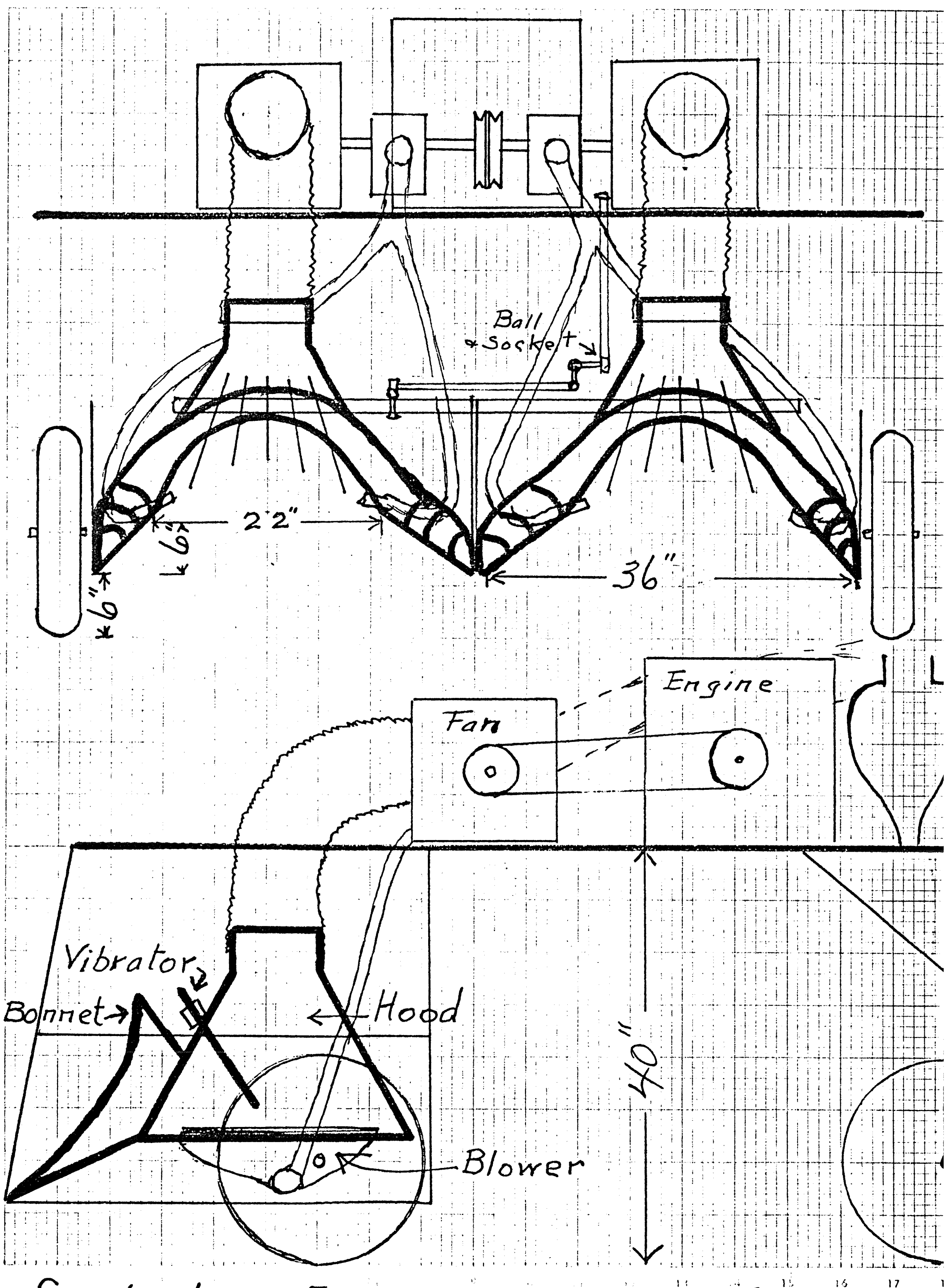
Power: One engine can be used to power the two-row machine, including perhaps hydraulic power "wheel motors" for propulsion, and for fans and vibrators.

Adjustable: The hood must be adjustable for height to harvest from different sized plants. It would also be desirable to have both the wheels and hoods adjustable for width to accommodate different widths of rows. (a 40 inch row seems desirable for preliminary seed production). The Denholm machine appeared to be fairly efficient,but it is heavy and expensive. The suggested machine should be light and not awfully expensive.

Seed container: Some recommend a "cyclone" which is good. However, we will use a large open mesh seed bag (part of it will be open mesh plastic, "Textaline", used in lawn furniture) for our first harvester since it is cheaper and lighter.

June, 1981

THE TYSDAL GUAYULE SEED HARVESTER



Scale 1cm = 5 inches

H.M.T.