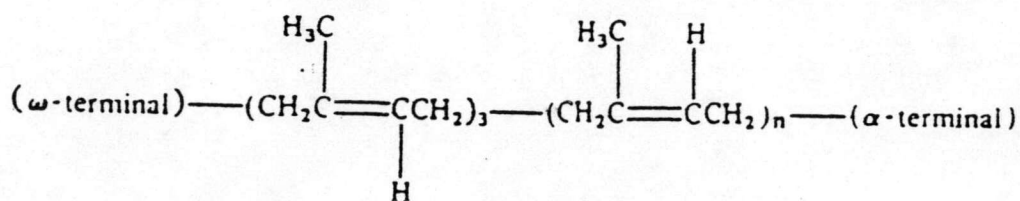# CHAPTER I

INTRODUCTION

Hevea <u>brasiliensis</u> or para rubber is the most important and commercially exploited plant for natural rubber production. In Thailand extensive cultivation of rubber in the Southern and Eastern parts are the main income of the people which also expand to the Northeast. Malaysia and Indonesia are the other two countries that export large amount of natural rubber.

The latex obtained from <u>Hevea</u> is a white or slightly yellowish opaque liquid. It is a weak lyophilic colloidal system of spherical or pearshaped rubber globules of size in the range 0.05-2.0 μm suspended in aqueous serum. The latex contains three main constituents : rubber (30-45%), non-rubber solids (3-5%) and water (55-67%). The main non-rubbers are proteins (2-2.5 g%), ash (0.7-0.9 g%), resin (1-1.6 g%), sugar (1.0-1.5 g%) and elements like Mg, Ca, Na etc. (Archer et al., 1963)

NMR spectroscopy (Tanaka, 1984) revealed the chemical structure of natural rubber as the polyisoprene with the building unit of isoprene (C_5H_8) in majority of cis-unit and approximately 2-3 tran-units per polymer chain as shown in Figure 1.1.

$$(\omega\text{-terminal})\!-\!(CH_2\overset{\overset{\displaystyle H_3C}{|}}{\underset{\underset{\displaystyle H}{|}}{C}}\!=\!=\!CCH_2)_3\!-\!(CH_2\overset{\overset{\displaystyle H_3C}{|}}{C}\!=\!=\!\overset{\overset{\displaystyle H}{|}}{C}CH_2)_n\!-\!(\alpha\text{-terminal})$$

The structure of both terminal units has not been identified.

Figure 1.1 The chemical structure of <u>Hevea</u> rubber

Commercially natural rubber (NR) can be classified into two major groups, solid NR and latex concentrate (Kajornchaiyakul, 1986). Solid NR has been produced from fresh field latex and divided into four groups, ribbed smoked sheet (RSS), air died sheet (ADS), crepe rubber and block rubber or Thai tested rubber (TTR), depending on its derived process. (Figure 1.2) In each group of solid rubber, the small number indicates for less impurities or better grade; such as TTR 5 has much less impurities than TTR 20, especially TTR 5L the letter "L" indicates for "light color grade", usually require bleaching.

All of these raw rubber materials are lined up to the rubber production process by which solid rubbers are transformed to various kinds of products. Rubber production process are composed of three main steps, compounding, molding and vulcanizing, in which the rubber materials are mixed with other chemicals or synthetic rubber to improve its physical properties as required. In order to achieve the highest
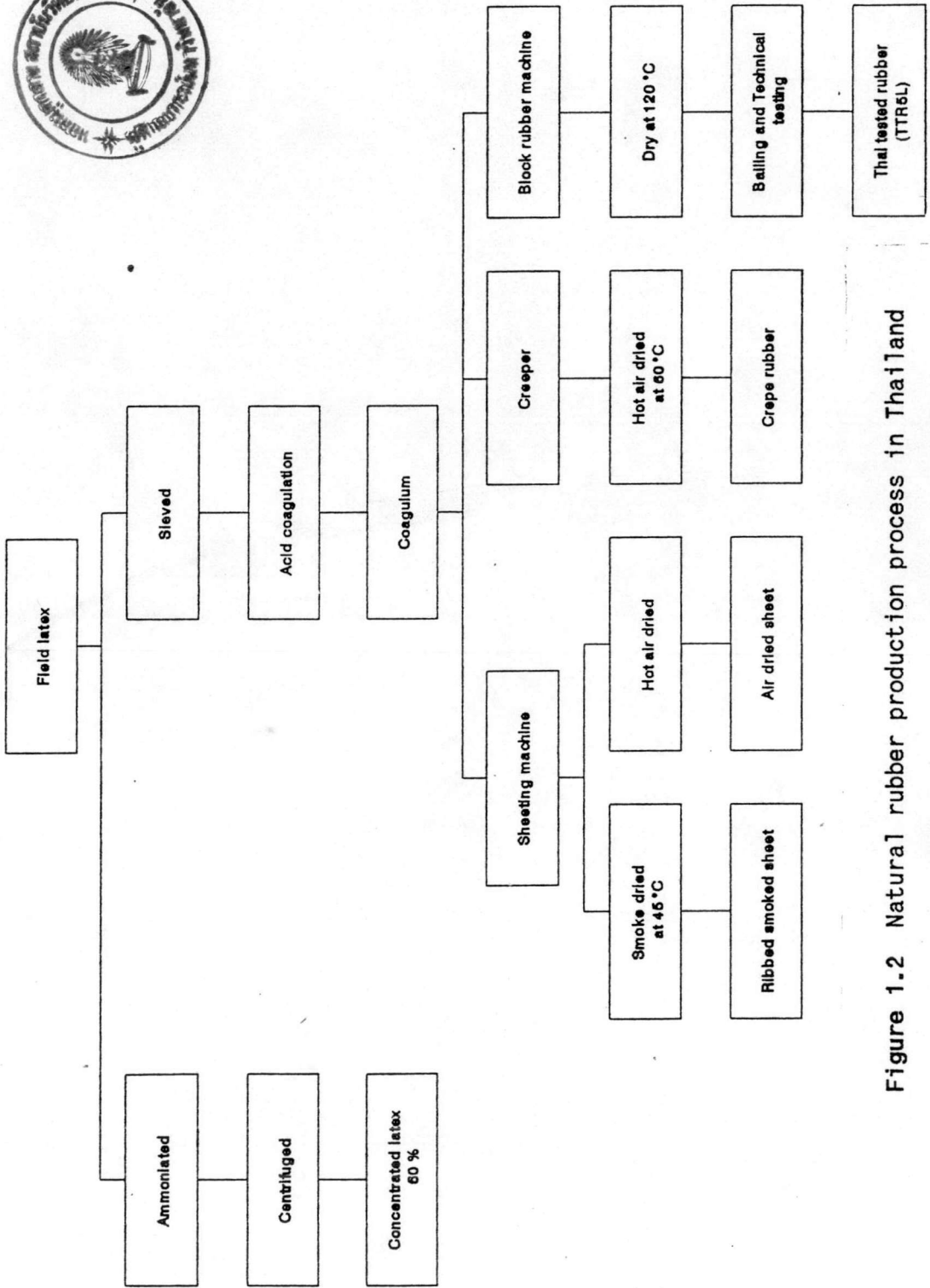
Figure 1.2 Natural rubber production process in Thailand

(From Para Rubber Bulletin, Thailand 7 (1986) : 89-98)

efficient rubber goods production which minimize the cost, loss and energy consumption. It should have the quality check in each step of the process and the quality of raw materials particularly the raw rubbers are the first important factor to determine not only the quality and consistency of rubber goods but also the processibility and productivity of the factory. (กฤษฎา สุชีวะ, 2531)

Such consistencies involve in the appearance of the rubber measured by visual inspection such as color, cleanness etc. and the rubber properties which required complicated techniques or instruments to analyze such as viscosity or plasticity and other non-rubber compositions.

Recently rubber manufactures are awared of the necessity to measure the viscosity of natural rubber lot by lot in order to adjust the mixing procedure accordingly (Long, 1985). Vershave (1988) representing Hutchinson, which is one of the world's major producers of industrial rubber goods has reported that the Mooney viscosity values of various grade of NR vary between 70-100 and is the most serious problem for rubber based industry. Hence, the developments of more uniform NR at a lower cost premium would obviously be advantageous for the producers and the understanding of the major causes of the variability must be an important step towards any improvement.

The bulk viscosity of rubber is an important property which is usually measured using either the Mooney viscosimeter or the Wallace plastimeter (Nair, 1970). Variation in the bulk viscosities of rubbers from different clone is one of the factors contributing to the reported inconsistency in the processing properties of NR. Such variation has

been shown to be dependent on the molecular parameters of the polymer, such as the molecular weight (MW) and molecular weight distribution (MWD), non-rubber constituents etc. and including the rubber processing. (Yip and Subramaniam, 1984).

Molecular weight (MW) and molecular weight distribution (MWD)
(Subramaniam, 1975)

The MW and MWD of NR are directly related to intrinsic viscosity and plasticity (Table 1.1) and there are a great variations in the average MW between rubbers of different clone although the range of MW of Hevea rubber always falls between 3×10^4 and 10^7, and the high molecular weight peak appear at $1-25 \times 10^6$ and the low molecular weight peak at $1-2 \times 10^5$ which can be classified in 3 different type of MWD as shown in Figure 1.3.

Table 1.1 Average molecular weights (MW), Wallace plasticity (Po), Mooney viscosity (MV), Molecular weight distribution (MWD) of some clonal crepe rubber (Subramaniam, 1975).

Clone	$M_W \times 10^{-6}$ from GPC	Initial plasticity (P_O)	Mooney viscosity ML 1+4(100 $^\circ$C)	Type of MWD
PB 5/51	2.18	67	94	3
PB 28/59	2.15	65	87	3
RRIM 600	1.93	53	72	2
GT 1	1.85	58	79	2

All samples were prepared by acid coagulation followed by creping any drying at 60°C.
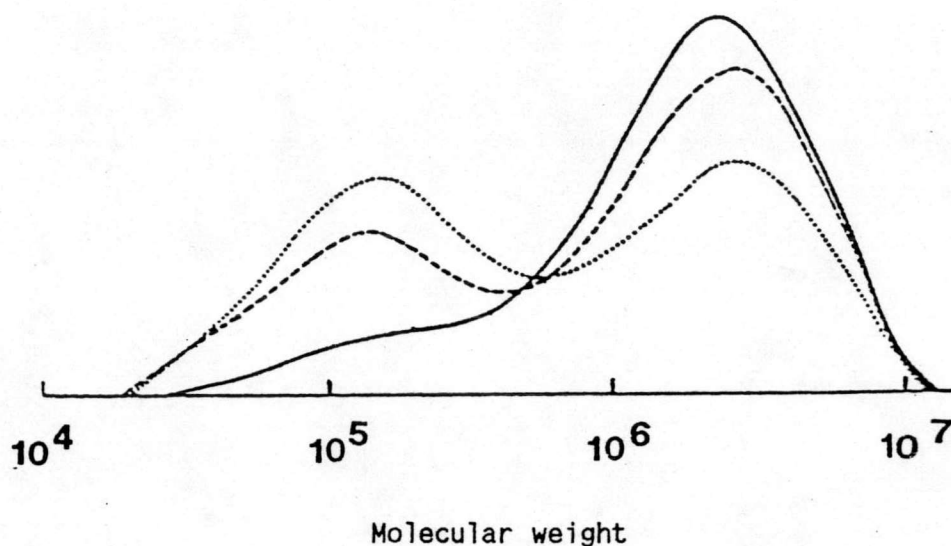
Molecular weight

Figure 1.3 Types of molecular weight distribution curves of natural rubber (Subramaniam, 1975)

Type 1. (....):Distinctly bimodal distribution where peaks are of nearly the same height.

Type 2. (----):Distinctly bimodal distribution where the peak in the low molecular weight'region is smaller.

Type 3. (_____):Skewed unimodal distribution with a "shoulder" or a "plateau" in the low molecular weight region.

Acetone extract or non-rubber content

Subramaniam (1975) has reported that total non-rubber content of rubber from different clones in Malaysia varies between 2-5% of total rubber content and broadly correlate with the Mooney viscosity and Wallace plasticity.

Gel phase

Hevea rubber contains 5-50% gel phase, which is insoluble in rubber solvents, depending on the clonal origin of the rubber, processing conditions and the time and temperature of storage (Tanaka, 1989). This phase consists of microgel and macrogel. While Freeman (1954); Allen and Bristow (1963) believed that the macrogel is caused by crosslinking between rubber molecules within the rubber particles, Wood (1952) believed that, the macrogel is associated with the abnormal groups distributing along the rubber chain, causing the storage hardening in dry rubber. However, the presence of gel correlates with an increase in the bulk viscosity of the rubber.

Storage hardening

The Mooney viscosity of bulk rubber tend to increase during manufacture and storage, causing the great variation in viscosity. Chin (1969) has demonstrated that there are great increase in Mooney viscosity of rubber during the process. After tapping, at that time the latex was added with ammonia until drying, each step has contributed to an increase in Mooney viscosity 5-8 units from its origin.

Such variation has been postulated to be mainly due to the crosslinking of special functional groups which are called abnormal groups (Sekhar, 1962). From the studies by infrared spectroscopy and recently ^{13}C-NMR spectroscopy (Tanaka, 1984) revealed the carbonyl or aldehyde groups along the polymer chain and certain aldehyde groups in the non-rubber phase e.g. amino acids are involved. This storage hardening is markedly-accelerated by conditions of low humidity at ambient temperature while it can be inhibited by the addition of amines such as hydroxylamine (Sekhar, 1961).

There are several factors causing discoloration of latex and of NR. As noted by Hasma & Subramaniam (1986), the color of latex depends on its clonal origin of rubber which related to its non-rubber compositions particularly pigments e.g. carotenoids etc. and its amount seem to vary among clones, climate and time of tapping. Moreover, the process of latex after tapping eg. ammonia addition acid coagulation and drying etc. also influence on NR color (Surasak Suttisonk,1986). However the most important factor causing discoloration of NR is generally believed as the enzymatic reaction, catalyzed by the polyphenol oxidase type (Nadarajah and Karunaratne, 1971) which are also responsible for the discoloration of many plant tissues. In latex, De Haan-Homans (1949) has reported that latex contains a complicated system of oxidizing enzymes, namely catalase, tyrosinase, peroxidase and other oxidases, especially in fluid of the lutoid particles known as B-serum (Southorn, 1966). When these particle are damaged, the enzymes move out and catalyze the oxidation of natural-occurring phenolic compounds which disperse in aqueous phase of latex or C-serum, enhanced by atmospheric oxygen to ortho-dihydroxy phenols and then to ortho-quinones, which react with naturally-occurring amino acids and proteins present in latex to give colored products resembling melanin. Reduced melanin is tan colors and oxidized melanin is black. Since latex exhibits gray, brown or black colors in discolored raw rubber, enzymatic oxidation should be one important factor. Discoloration is also caused by non-enzymatic reaction between carbonyl and amino groups to give an unsaturated carbonyl amino derivative, showing brown or black colors (Rinderknecht and Jurd, 1958).

Since Oxygen is essential for the oxidative formation of discolored products, sodium bisulfite or sodium metabisulfite, a strongly reducing agent, is used as inhibitor either by removing O_2 from reaction or by reducing highly reactive O-quinones to polyphenols (Burton et al., 1963). The normal dosage of Sodium metabisulfite is 0.03 - 0.05% based on dried rubber content (DRC).

Protein and its effects

Among non-rubber constituents, proteins are major components. About 20% of total proteins in the latex are absorbed on the surface of rubber particles, and the same amounts are associated with the bottom fraction (or B-serum), obtained by high speed centrifugation. The remainders are dissolved in the aqueous phase of latex or C-serum (Archer, 1963). From various types of electrophoresis it has been shown that C-serum proteins consist about 19 anionic and 5 cationic proteins (Tata and Edwin, 1970) of which α-globulin, isolated by Archer and Cockbain (1955), forms the major protein. The proteins in the bottom fraction consist of 8 anionic a 5 cationic proteins (Karunakaran et al., 1961) in which hevein and hevamine have been isolated (Archer, 1960; Archer, 1976).

However, there are many more kinds of protein distributed in different phase of Hevea latex which should have been studied in its properties and effects on rubber polymers because only minute amounts of protein can cause many deleterious effects on rubber properties.

Proteins, by their polar and hydrophilic nature, are believed to affect the properties of rubber in many aspects from the raw rubber properties to the vulcanizate properties. It is believed to involve in

the hardening phenomenon of bulk rubber which Tanaka (1984) has shown that newly-formed network, containing a larger proportion of nitrogen content, may be occurred by the formation of protein with other particles. Borgotrom (1968) also believed that enzyme polyphenol oxidase (PPO) is in charge of latex or NR discoloration.

During processing, protein has been suggested to act as filler, having variable stiffening effect, resulting in poor dispersion of vulcanized curatives and eventually leading to local over-crosslinking and properties variation (Bloomfield, 1973). In a later contribution Metherell (1980) has shown that stiffening action is one source of modulus variation because it is sensitive to moisture and is reduced at high relative humidity. Water absorbed by protein and other hydrophilic non-rubbers can increase stress relaxation and crepe or easily deformed under loading as a result of a reduction in modulus. Absorption of water by unvulcanized rubber can also affect the degree of crosslinking and other cure characteristics of some vulcanizing system, thus given rise to a further source of modulus variation.(Elliott et al.,1970)

To overcome these problems, there are several attempts to produce a rubber with a very low protein content in order to improve a rubber properties. This rubber with a low protein or nitrogen content has been generally known as "Deproteinized Natural Rubber", DPNR or "Low Nitrogen Natural Rubber", LNNR. The rubber compositions of DPNR produced by Rubber Research Institute of Malaysia (RRIM) since 1977 has been specified as shown in Table 1.2.

Table 1.2 Typical properties of DPNR

Property	DPNR from Latex concentrate	DPNR from clarified field latex	Proposed specification
Nitrogen content(g%)	0.066	0.12	less than 0.15
Dirt (g%)	0.006	0.005	less than 0.015
Ash (g%)	0.052	0.13	less than 0.15
Volatile matter (g%)	0.28	0.25	less than 0.5
Initial plasticity (P_o)	39	32	-
Plasticity Retention Index (PRI)	66	85	60 (min.)
ΔP (P_H-P_o)	-	7	No value > 9
Mooney viscosity ML 1+4 (100 ^oC)	-	51	45-55, 55-65

DPNR is very closely similar in physical and chemical characteristics to synthetic cis 1,4-polyisoprene, it has better green strength and building tack than its synthetic analogue. When compounded with soluble vulcanizing ingredients, DPNR exhibits improved lower creep and compression set properties than those of conventional grades of natural rubber (Bloomfeild, 1973). It has also excellent technological and dynamic properties such as low heat build up (Knight and Tan, 1975), low water or sea water absorption (Connolly et al., 1970), superior fatigue life and uniformity in cure behavior (Bernard, 1973). All these features are well exploited when DPNR is used for engineering application .

Several methods that have been used for removing latex protein from different types of latex are summarized in Table 1.3.

Table 1.3 Developments in the manufacture of DPNR

Starting material	Removing agent	Reference
Skim latex	Sodium hydroxide	Firestone Tyre rubber Co., 1955
Field latex	Dioctyl - octyl sodium sulfosuccinate	John, 1971
Skim latex	Sodium sulfosuccinate and calcium chloride	John and Sin, 1973
Field latex	Papain	Nadarajah et al., 1973
Latex concentrate	Superase	Chin et al., 1974
Skim latex	Trypsin	Ong, 1974
Skim crumbs	Sodium hydroxide and oxalic acid	Ong, 1974
Field latex	Papain/$NH_2OH.HCl$	Yapa, 1975
Clarified latex	Superase or Alcalase	Chang et al., 1977
Clarified latex	Superase /$NH_2OH.HCl$	Chang et al., 1977
Field latex	Papain/Alkaline treatment	Yapa, 1977
Field latex	Papain/Alkaline treatment/ $NH_2OH.HCl$	Yapa, 1977
Field latex	BPN/Alkaline treatment/ $NH_2OH.HCl$	Yapa, 1977
Field latex	Superase/Alkaline treatment/ $NH_2OH.HCl$	Yapa, 1977
Skim latex	Papain	Yapa et al., 1978
Field latex	Pineapple juice (bromelain)	Yapa et al., 1980

In summary, these are three main methods to remove proteins.

1) By treatment with some kinds of surfactant or detergent by which proteins are eluted from the rubber surface.

2) By chemical treatment such as soaking with NaOH for 24 h by which proteins are hydrolyzed by chemical reaction.

3) By biochemical treatment with proteolytic enzymes such as papain, trypsin, Alcalase etc. which hydrolyze proteins into small peptides and amino acids which are more soluble for washing out. The deproteinization scheme can be carried out by combination of these treatments in order to remove proteins out as much as possible. However, enzyme treatment is the most useful method because it can be carried out in mild condition and quite specific with less side effect on rubber molecules.

DPNR production and sale

Figure 1.4 show the current strategic of DPNR production and sales which is quoted from RRIM station between 1985-1989 with the short remarks on its developments.

In 1985-1986, the initial stage of DPNR factory scale production, DPNR was firstly introduced since 1974 in RRIM laboratory. By earlier process, DPNR was too expensive to use in any application and many users may not be awared of the benefits of using them. In 1987, the new factory-scale process was developed for DPNR production. Unlike the earlier process, no clarification or dilution of the latex is required and coagulation is by steam in a column coagulator. The new process is cheaper than the earlier process. Moreover, the new DPNR has lower nitrogen and Volatile matter values and light color. However, it
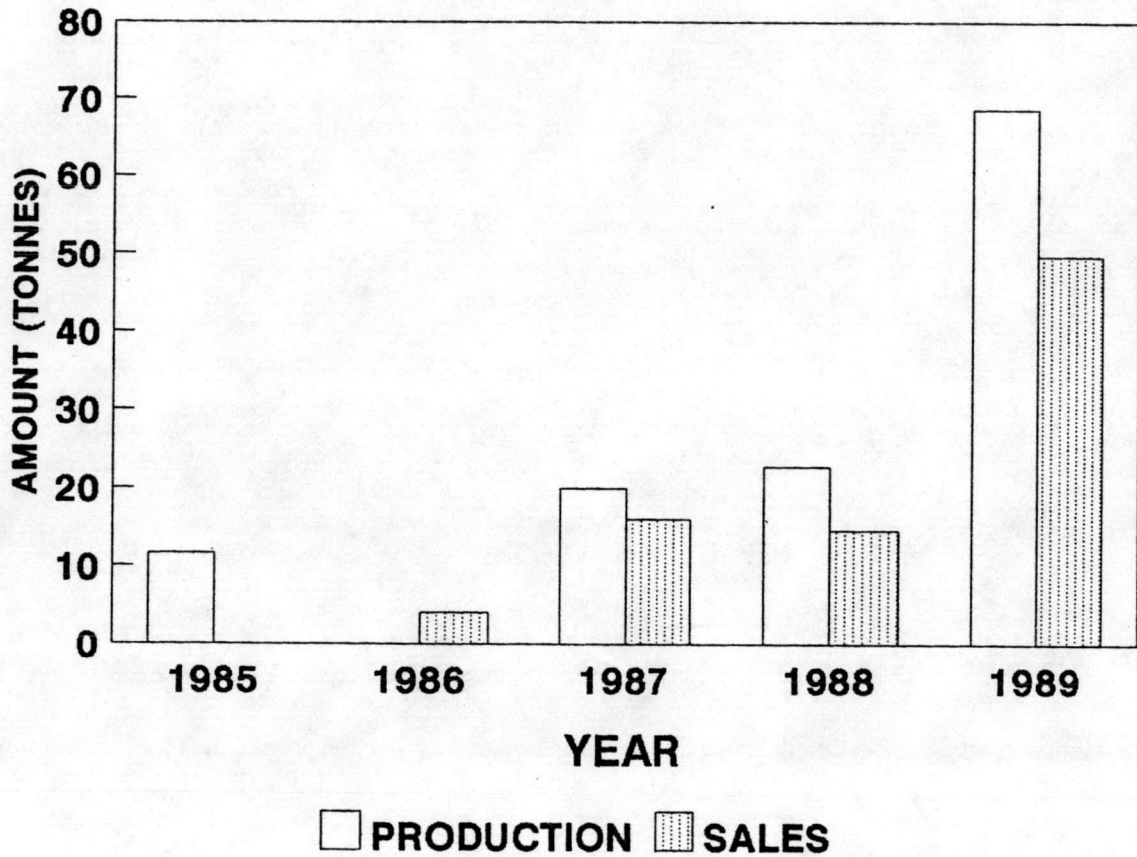
Figure 1.4   DPNR production and sales

(From RRIM Planter's Bulletin 1985-1989)

is relatively more scorchy. In 1988, Further improvements were made to the "new" DPNR process including transformation of the batch process into a continuous one.   This improved consistency and slightly reduced production cost.   In 1989, Further improvements were made to the DPNR process line.   However, the scorch problem remained unsolved.   After introduction of an improved DPNR in 1987, the production and sales for DPNR is projected to reach 70 tones in 1990, from an almost non-existent demand in 1986.   Indications are that demand will continue to rise.

For Thailand, the country has been ranked the Third among the world rubber producers and will be the First in the this decade. However, there are very few studies and developments of new rubber production which will lead to the production of highly qualified form of more value-added rubber goods. The purpose of this study is first of all to optimize the condition for CV-DPNR production by two locally available enzymes, Papain and Alcalase, from both starting materials : latex concentrate 60%, and clonal fresh field latex. Secondly to study the physical properties of CV-DPNR produced under these optimized condition, and compare with control non deproteinized and the best commercially available NR grade, TTR 5L and crepe rubber.