# PHYSICO-CHEMICAL PROPERTIES OF STARCH IN SAGO PALM (Metroxylon sagu) AT DIFFERENT GROWTH STAGES

# ADRINA TIE PEI LANG

UNIVERSITI SAINS MALAYSIA 2004

# PHYSICO-CHEMICAL PROPERTIES OF STARCH IN SAGO PALM (Metroxylon sagu) AT DIFFERENT GROWTH STAGES

by

# ADRINA TIE PEI LANG

Thesis submitted in fulfillment of the requirements for the degree of Master of Science

August 2004

#### **ACKNOWLEDGEMENTS**

And finally, IT IS ACCOMPLISHED! Praise be to GOD for seeing me through it all.

I wish to express my heartfelt thanks to my two very hardworking and dedicated supervisors, Dr Abdul Manan Dos Mohamed and Associate Professor Dr Abd. Karim Alias for their guidance, motivations and optimistic outlook in the course of my research and thesis writing.

My earnest thanks also to YBhg. Dato' Dr Abdul Halim bin Hj Mohd Hassan, Datin Dr Zaliha Christine Abdullah and Dr Noraini Busri who helped to conceive the concept behind this study.

I wish also to share this achievement with my fellow colleagues (in Kuching and Mukah) of CRAUN Research Sdn. Bhd. headed by YBhg. Encik Yusup Sobeng. My sincere gratitude for the use of the facilities, the cooperation and assistance rendered in one way or another. I would especially like to thank Mr Peter Mittis for his commitment and diligence in assisting me with my research work.

My appreciation and thanks to Dr Yusrida Darwis (Pusat Pengajian Sains Farmasi) for assistance with Malvern Mastersizer, to Kak Jamilah (Pusat Pengajian Kaji Hayat) for help with SEM, to the staff and my fellow post-graduate mates of Pusat Pengajian Teknologi Industri for their kind generosity and hospitality during my attachment in USM.

As I reflected back, I am grateful to my family for keeping me in touch with LIFE apart from work. In my anxiety to finish my MSc, the presence of Ma, Pa, Ah Lek, Ah Wei, Julia, Ah Woo, Mee King and baby Jia Yi has added some spices and colours to my life, reminding me to keep a balance in living each day, to the fullest.

Last but not least, I thank God for the many dear friends who are my companions on the journey. In my many moments of disappointment and dismay, the thoughts of your prayers and moral support have helped me to keep my chin up and journey on in faith and hope. Thank you for being my sheltering trees.

## TABLE OF CONTENTS

		page
	ACKNOWLEDGEMENTS	ii
	LIST OF TABLES	vii
	LIST OF FIGURES	ix
	ABSTRACT	xi
	ABSTRAK	xiii
1	INTRODUCTION	1
	1.1 Background	1
	1.2 Specific Objectives	4
	1.3 Research Protocol	4
2	LITERATURE REVIEW	5
	2.1 The Sago Palm	5
	2.1.1 Taxanomy	5
	2.1.2 Historical Origin and Distribution	5
	2.1.3 Botany	7
	2.1.4 Extraction of Sago Starch	17
	2.1.4.1 Traditional Method of Extraction	17
	2.1.4.2 Modern Method of Extraction	18
	2.1.5 Quality of Sago Starch	19
	2.1.5.1 Industrial Grade Sago Starch	19
	2.1.5.2 Edible Grade Sago Starch	19
	2.1.6 Utilisation of Sago Starch	20
	2.1.6.1 Traditional Uses	20
	2 1 6 2 Uses in Food Industries	21

		2.	1.6.3	Uses in Non-Food Industries	2	2
		2.	1.6.4	Uses in Biotechnology	2	22
		2.	1.6.5	Other Industries	2	22
	2.2	Sta	rch		2	23
		2.2.1	Amy	lose and Amylopectin	2	23
		2.2.2	Struc	ture and Organisation in Starch Granules	2	28
		2.2.3	Varie	eties of Starch	2	29
		2.2.4	Swell	ling and Gelatinisation	3	3(
		2.2.5	Therr	mal Analysis of Starch Gelatinisation	3	32
		2.2.6	Retro	ogradation	3	35
		2.2.7	Flow	Behaviour	3	37
		2.	2.7.1	Principles of Flow Measurement	2	4(
		2.2.8	Partic	cle Size Analysis	2	42
		2.	2.8.1	Principles of Particles Size Analysis	2	45
	2.3	Ass	ociate	d Components in Sago Pith	2	47
		2.3.1	Crud	e Fibre and Cellular Materials	2	47
		2.3.2	Pheno	olic Compounds in the Pith	4	49
3	MA	TERI	ALS &	z METHODS	5	53
	3.1	Loc	ation (	of Sago Palm Plot	5	53
	3.2	San	npling	of Sago Palms	5	53
	3.3	San	nple P	reparation	5	56
	3.4	Rea	gents	and Chemicals	5	56
	3.5	Det	ermin	ation of Moisture Content	5	56
	3.6	Ext	ractio	n of Sago Starch	5	58
	3.7	Ana	alysis o	of Pith Tissue	5	58
		3.7.1	Deter	rmination of Total Starch Content	5	58
		3.7.2	Morp	phological Studies	6	52
	3.8	Cha	aractei	risation of Physico-Chemical Properties	6	52
		3.8.1	Deter	rmination of Swelling Factor	$\epsilon$	52
		3.8.2	Partic	ele Size and Distribution Analysis	$\epsilon$	53
		3.8.3	Flow	Behaviour Measurement	$\epsilon$	53
		3.8.4	Pastii	ng Profile Analysis	$\epsilon$	54
		3.8.5	Therr	mal Profile Analysis	6	55
		3.8.6	Retro	ogradation Analysis	$\epsilon$	56
	3.9	Cor	nposit	ion of Associated Components in Sago Pith	6	56
	3.9.1 Determination of Soluble Carbohydrate					56

	3.9.2	2 Determination of Phenolic Compound	67
	3.9.3	Determination of Lignin Content	68
	3.9.4	Determination of Non-Starch Polysaccharides (NSP)	69
	3.10 Sta	atistical Analysis	74
4	RESULT	CS AND DISCUSSION	75
	4.1 Gr	owth Data and Estimated Age	75
	4.2 Ar	nalysis of Pith Tissue	77
	4.2.1	Total Starch Content	77
	4.2.2	2. Moisture Content	80
	4.2.3	Morphological Studies	82
	4	4.2.3.1 Sago Starch	82
	4	4.2.3.2 Sago Hampas	84
	4.3 Ch	naracterisation of Physico-Chemical Properties	86
	4.3.1	Swelling Factor	86
	4.3.2	Particle Size and Distribution Analysis	89
	4.3.3	Flow Behaviour Measurement	93
	4.3.4	Pasting Profile Analysis	97
	4.3.5	Thermal Profile Analysis	108
	4.3.6	6 Retrogradation Analysis	119
	4.4 Co	omposition of Associated Components in Sago Pith	126
	4.4.1	Total Soluble Carbohydrate	126
	4.4.2	2 Total Phenolic Compound	128
	4.4.3	3 Total Lignin Content	130
	4.4.4	Non-Starch Polysaccharides Analysis	131
5	CONCL	USIONS	136
6	FUTURE	ERESEARCH	138
7	REFERE	ENCES	139
8	APPEND	DICES	
	A Pr	eparation of Nelson-Somogyi Reagent Mixture	159
	B Pr	eparation of Nelson Arsenomolybdate Reagent	160
	C Gl	ucose Standard Curve	161
	D Pr	eparation of Folin-Denis Reagent	162
	E Ph	enolic Compound Standard Curve	163

F	Preparation of Enzyme 1 & 2 (NSP)	164
G	Chromatogram of Non-Starch Polysaccharides Sugar	165
	Standards	
Н	Uronic Acid Standard Curve	166
I	Statistical Analysis using ANOVA (Microsoft® EXCEL 2002)	167
	at $\alpha = 0.05$ (95 %) confidence level	
J	Growth Parameters of Sago Palms	168
K	<b>Total Starch and Moisture Content</b>	173
L	Swelling Factor	174
M	Particle Size and Distribution	175
N	Flow Behaviour Measurement	176
P	Pasting Profile	177
Q	Thermal Profile	178
R	Retrogradation Profile	179
S	Total Soluble Carbohydrate	180
T	Total Phenolic Compound and Lignin Content	181
U	Non-Starch Polysaccharides	182
V	Chromatogram of Neutral Sugars from Sago Pith Samples	184
W	Papers arising from the project	185

## LIST OF TABLES

Table	Title	Page
2.1	Distribution of sago palm areas (Tie et al., 1991)	6
2.2	PELITA Sago Estate Development (PELITA, 2003)	7
2.3	Different physiological growth stages of sago palm (Lim, 1991a)	10
2.4	Stages of growth and development of sago palm in Sarawak (Jong, 1995)	11
2.5	Requirements for Industrial sago starch (MS468, 1976)	19
2.6	Requirements for Edible sago starch (MS470, 1992)	20
2.7	Uses of sago flour in small industries (Sim, 1986)	21
2.8	Crystal polymorph and amylose content of various starches (Oates, 1997)	28
2.9	Granule properties of various starches (Swinkels, 1990)	30
2.10	Proximate analyses of coarse and fine hampas (Cecil <i>et al.</i> , 1982)	48
3.1	Standard measurement cycle (RVA Standard 1)	65
4.1	Growth data and estimated age of sago palms at different growth stages	76
4.2	Comparison of mean total starch and moisture content of sago core (bore) pith and section pith samples at base and mid heights of the different growth stages	78
4.3	The swelling factors (at 70 °C) of sago starch from base and mid heights of different growth stages	87
4.4	Particle size and distribution profile of sago starch from base and mid heights of different growth stages measured by the Malvern Mastersizer	90
4.5	Flow behaviour measurement of sago starch (4 % w/w slurry) at base (B) and mid (M) heights of palms at different growth stages	95
4.6	The pasting profile of sago starch at base and mid heights of different growth stages determined by the Rapid Visco Analyser	100

Table	Title	Page
4.7	Thermal properties of sago starch:water (1:1) system from base and mid heights at different growth stages determined by Differential Scanning Calorimetry	111
4.8	Thermal profile of retrograded starch (starch:water at 1:1, kept at 5 °C for 4 days) from base and mid heights of different growth stages determined by Differential Scanning Calorimetry	121
4.9	Comparison of total soluble carbohydrate, total phenolic compound and total lignin content with total starch content in the sago pith from base and mid heights of the different growth stages	127
4.10	Total, insoluble and soluble non-starch polysaccharides in sago pith of base and mid heights at different growth stages	132

## LIST OF FIGURES

Figure	Title	Page
2.1	Life cycle of a sago palm (Schuiling & Flach, 1985)	9
2.2	Plawei – palm that has reached maximum vegetative growth	12
2.3	Plawei Manit – inflorescence emerging palm	13
2.4	Bubul – inflorescence developing palm	14
2.5	Angau Muda – flowering palm	15
2.6	Angau Tua – fruiting palm	16
2.7	Linear and branched starch polymers (Murphy, 2000)	24
2.8	Model of amylopectin structure according to Hizukuri (1986)	26
2.9	Schematic diagram of the structure of a starch granule. a), single granule comprising concentric rings, each containing stacks of amorphous and crystalline lamellae; b) amorphous and crystalline lamellae; c) chains of amylopectin arranged in a cluster structure (Jenkins & Donald, 1995)	27
2.10	Differential scanning profiles of potato starch at various solvent fractions (a volume of 0.6 corresponds to about 50 %). The heating rate was 10 °C min <sup>-1</sup> (Donovan, 1979)	34
2.11	Rheogram showing Newtonian and Non-Newtonian flow (Holcomb & Tung, 1991)	39
2.12	Velocity profile for an ideal viscous fluid in steady laminar shearing flow between two parallel plates (Holcomb & Tung, 1991)	41
2.13	Normal or Gaussian Distribution	46
2.14	Bimodal Distribution	46
2.15	DL-epicatechin (Ozawa & Arai, 1986)	51
2.16	D-catechin (Ozawa & Arai, 1986)	51
2.17	(2S)-4'-hydroxy-5,7-dimethoxy flavane (Ozawa & Arai, 1986)	51
2.18	(2S)-5-methoxy-7-hydroxy flavane (Ozawa & Arai, 1986)	51

Figure	Title	Page		
3.1	Location of Sungai Talau Peat Research Station	54		
3.2	Data collection and sampling of sago palm	55		
3.3	Sampling using the increment borer tool (Jozsa, 1988)	57		
4.1	The moisture and total starch content of core pith samples from base (B) and mid (M) heights of the different growth stages	81		
4.2	Scanning electron micrographs of sago starch granules (200 x magnifications) extracted from base height of (a) Plawei; (b) Angau Muda; (c) Angau Tua; and (d) Late Angau Tua stages.	83		
4.3	Scanning electron micrographs of sago hampas (150 x magnifications) from (a) Bubul; (b) Angau Muda; and (c) Angau Tua stages.	85		
4.4	Swelling factor of sago starch from base (left) and mid (right) heights of different growth stages	88		
4.5	Particle size distribution pattern of sago starch at base and mid heights of different growth stages	92		
4.6	Flow behaviour measurement of sago starch at base (left) and mid (right) heights of the different growth stages	94		
4.7	A typical pasting curve of sago starch			
4.8	Pasting curves of sago starch at base (left) and mid (right) height of different growth stages determined using the Rapid Visco Analyser			
4.9	Pasting properties of sago starch at base and mid heights of different growth stages	104		
4.10	A typical DSC thermogram of sago starch:water (1:1) system scanned at 10 °C min <sup>-1</sup>	109		
4.11	DSC thermograms of 1:1 sago starch:water systems at base (left) and mid (right) heights of palms at different growth stages	113		
4.12	Thermal profile and enthalpy of gelatinisation of sago starch at base and mid heights of different growth stages	114		
4.13	The range of sago starch at base and mid heights of different growth stages in comparison to the transition temperatures			
4.14	DSC thermograms of 1:1 retrograded sago starch:water systems at base (left) and mid (right) heights of palms at different growth stages	120		
4.15	Retrogradation properties of sago starch at base and mid heights of different growth stages	124		
4.16	Composition of associated components in the sago pith	129		
4.17	Non-starch polysaccharides in comparison with total starch content at base and mid height of different growth stages	135		

#### **ABSTRACT**

This study aimed to characterise the physico-chemical properties of sago starch from base and mid heights of palms at different growth stages, namely, 'Plawei', 'Bubul', 'Angau Muda', 'Angau Tua' and 'Late Angau Tua'. The starch content and the composition of associated components in the sago pith were determined as well. The characterisation of physico-chemical properties of sago starch involved determination of swelling factor, particle size and distribution profile, flow behaviour of the starch paste, pasting characteristics and retrogradation profiles analysis. The starch content was found to increase as the palms matured from Plawei to Angau Muda stages and decrease from Angau Tua to Late Angau Tua stages. The scanning electron micrographs of sago starch showed oval-shaped granules of 10 - 30 µm in different proportion. Variation was observed in the granule size of starch whereby the starch at base height of all the stages was larger in mean diameter than mid height. The pasting profile showed four different pattern of pasting curves from the combination of the mean results. No prominent variation was observed in the results of swelling factor, flow behaviour measurement and thermal properties of sago starch from the different growth stages. All the starch samples showed the highest swelling factor at 70 °C. The best fit curve of shear stress versus shear rate indicated all the samples fitted the Herschel-Bulkley model. The relationship between the log viscosity and log shear rate suggested that the starch dispersion is susceptible to shear-thinning or pseudoplastics behaviour. The thermal

profile showed similar thermograms with  $T_0$  in the range of 70.2 - 73.1 °C,  $T_p$  of 75.1 - 77.0 °C,  $T_c$  of 97.4 - 101.4 °C and  $\Delta H_G$  of 17.5 - 19.2 Jg<sup>-1</sup> in all the starch samples. Similarly, the thermograms of retrograded starch showed a broad endotherm occurring at transition temperatures of 25 - 29 °C and  $\Delta H_R$  of 8 - 9 Jg<sup>-1</sup> lower than that of gelatinised starch. No significant difference was observed in the composition of associated components of sago pith from the different growth stages. The soluble carbohydrate ranged from 4.5 - 8.5 %. The phenolic compound was less than 1 % whereas the lignin content ranged from 9 - 22 %. The non-starch polysaccharides in the form of total, insoluble and soluble non-starch polysaccharides ranged from 57.5 - 105.4 %, 43.5 - 86.4 % and 12.4 - 23.9 %, respectively. In conclusion, the best stage for harvesting is Angau Muda stage whereas the variation in the physico-chemical properties of sago starch from base and mid heights of different stages will govern its application in the different industries according to the individual commercial needs.

# SIFAT-SIFAT FIZIKO-KIMIA KANJI DALAM PALMA SAGU (Metroxylon sagu) PADA PERINGKAT PERTUMBUHAN YANG BERBEZA

#### **ABSTRAK**

Kajian ini bertujuan untuk menentu sifat-sifat fiziko-kimia kanji sagu di ketinggian dasar dan tengah palma dari peringkat pertumbuhan yang berbeza, iaitu 'Plawei', 'Bubul', 'Angau Muda', 'Angau Tua' dan 'Late Angau Tua'. Kandungan kanji dan komposisi komponen-komponen berkaitan dalam empulur sagu juga ditentukan. Penentuan sifat-sifat fiziko-kimia kanji sagu merangkumi analisis-analisis faktor pembengkakan, saiz granul dan profil taburan, aliran kelakuan pes kanji, sifat pempesan dan profil retrogradasi. Kandungan kanji didapati meningkat apabila palma matang dari peringkat 'Plawei' ke 'Angau Muda' dan menurun dari peringkat 'Angau Tua' ke 'Late Angau Tua'. Mikrograf penskanan elektron menunjukkan granul sagu berbentuk lonjong dan mempunyai saiz 10 – 30 micron dalam perkadaran yang berbeza. Variasi didapati dalam saiz granul kanji di mana purata diameter granul kanji di ketinggian dasar pada semua peringkat pertumbuhan adalah lebih besar daripada ketinggian pertengahan. Kombinasi keputusan min profil pempesan menunjuk empat lengkungan yang berlainan corak. Tiada variasi yang ketara diperhatikan dalam keputusan faktor pembengkakan, kelakuan aliran dan sifat termal kanji sagu dari semua peringkat pertumbuhan yang berlainan. Kesemua sampel-sampel kanji menunjuk factor pembengkakan tertinggi pada suhu 70 °C. Lengkungan 'shear stress' lawan 'shear rate'

menunjukkan kesemua sampel mematuhi model 'Herschey-Bulkley'. Hubungan 'log viscosity' dan 'log shear rate' mencadangkan bahawa ampaian kanji mudah mengalami 'shear-thinning' atau 'sifat 'pseudoplastic'. Profil termal menunjukkan termogram yang serupa dengan  $T_0$  dalam linkungan 70.2 - 73.1 °C,  $T_p$  75.1 - 77.0 °C,  $T_c$  97.4 - 101.4 $^{\circ}\text{C}$  dan  $\Delta H_{G}$  17.5 - 19.2  $\text{Jg}^{\text{-1}}$  untuk kesemua sampel. Termogram untuk kanji retrogradasi menunjukkan satu endoterma yang lebar pada suhu peralihan 25 – 29 °C dan ΔH<sub>R</sub> 8 - 9 Jg<sup>-1</sup> kurang daripada kanji yang tergelatinisasi. Tiada perbezaan yang ketara diperhatikan dalam komposisi komponen-komponen berkaitan dalam empulur sagu pada peringkat pertumbuhan yang berlainan. Amaun karbohidrat terlarut adalah dalam banjaran 4.5 – 8.5 %. Kompaun fenolik adalah kurang daripada 1 % manakala kandungan lignin adalah 9-22 %. Polisakarida bukan-kanji dalam bentuk total, tidak larut dan larut adalah masing-masing 57.5 - 105.4 %, 43.5 - 86.4 % and 12.4 - 23.9 %. Sebagai kesimpulan, peringkat pertumbuhan terbaik untuk ditebang adalah 'Angau Muda' manakala variasi dalam sifat-sifat fiziko-kimia kanji sagu dari ketinggian dasar dan tengah pada peringkat pertumbuhan berlainan akan menentukan aplikasi dan penggunaan kanji sagu dalam industri-industri yang berlainan, bergantung kepada keperluan komersial individu.

#### 1 INTRODUCTION

#### 1.1 Background

Sago palm (*Metroxylon spp.*) is one of the few tropical crops which can tolerate wet growing conditions including peat swamps (Jong, 1995). Sago palm is also one of the oldest tropical plants exploited by man for its stem starch (Mathur *et al.*, 1998). Since the 1900s, much study has been carried out on sago palm cultivation. These included Nicholson (1921), Salverda (1947), Vegter *et al.* (1983) as cited by Flach in 1984. In the 1970s, much attention was concentrated on studies of sago palms in Sarawak where export of sago flour was fast becoming one of the important agricultural export commodities, with export of about 28 thousand tonnes of industrial grade sago starch earning about RM3.8 million in 1970 to about RM8.8 million for about 26 thousand tonnes in 1980. In 2002, export of about 34.6 thousand tonnes of food grade sago starch earned about RM28 million (Department of Statistic Malaysia, Sarawak Branch, 2003).

Sago palm has a main advantage of the ability to thrive in the harsh swampy peat environment (Ruddle, 1977; Johnson, 1977) which covers an area of 1.5 million ha i.e., 12 % of Sarawak's total land area (Tie & Lim, 1977). Based on this fact, the Sarawak Government has intensified their effort to further develop the sago industry through the Department of Agriculture, Sarawak (1982). Further to this, a commercial sago plantation (1987) was developed in Mukah by the Sarawak land development agency (Land Custody and Development Authority, LCDA) as well as a crop research and development unit (1993) to undertake more intensive research and development on sago (Jong, 1995).

The sago palm (*Metroxylon spp.*) is exploited both as a staple and cash crop in Sarawak. In its wild, semi-wild and cultivated forms, it is found throughout the coastal belt of Sarawak, but is concentrated in the rivers areas in Mukah District, Third Division (Kueh, 1977). Sago starch accumulates in the pith core of the stem of the sago palm (Cecil *et al*, 1982). The starch reserves are apparently at their maximum just before flowering, and fruiting deplete these reserves (Ruddle *et al.*, 1978). In Indonesia and Sarawak, the general belief is that the felling of sago palm is best carried out after flowering but before the fruiting stage (Tan, 1982). Traditionally, the starch is extracted manually by shredding the pith using an adze tipped with a hard wood blade. The shredded pith is trampled on a platform where water is added and starch was allowed to pass through finely-woven reed mat. The starch slurry collected was allowed to settle and after draining the water, the solid wet flour (*lamentak*) was spread on mats to dry in the sun (Morris, 1977).

In the modern factories however, the logs on arrival at the mills are immediately processed by first being debarked, followed by maceration using a rasper. The hammer mill further disintegrated the rasped pith into finer pieces and the starch slurry was passed through a series of centrifugal sieves and cyclone separators. The semi-dried starch from the rotary vacuum drum dryer is further dried by hot air drying in the flash dryer (Azudin & Lim, 1991).

The different methods of starch extraction gave rise to various quality of sago starch. The early European and Chinese entrepreneurs who set up businesses in Kuching or Singapore preferred to buy crude wet flour and refine it in their own factories. Thereon, the sago trade had become very valuable since the establishment of the international market for cheap industrial starch in Singapore (Morris, 1977). Attempts at improving the quality of sago flour exported in Sarawak was carried out by

the Colonial Government. In the 1950s, a Sago Advisory Board was formed which set the minimum standard based on appearance (colour) and the amount of fibre (Ong, 1977). In the decades that followed, the standard was improved by the Standards and Industrial Research Institute of Malaysia (SIRIM) stating the requirements for industrial grade sago starch (MS468 in 1976) and edible food grade sago starch (MS470 in 1992).

As sago starch grew in popularity as an alternate source of starch which is cheaper in price in the 1980s, many consumers and end users in the food industries started replacing other starches such as tapioca and corn, with sago starch. However, these industries soon faced problems/disadvantages of using sago starch, such as, the distinct sago smell and lack of protein fortification in bread-making (Clarke *et al.*, 1977). In another case, Müller found 3 limiting factors in replacement of maize and other cereals with sago flour in poultry and pig diets namely, (i) the inconsistent quality grade of sago, (ii) the balancing of nutrients in formulation of sago-based diets, & (iii) the volume and fine texture of sago-based diets (Müller, 1977).

In the Japanese researchers' studies on improvement of sago starch quality, Fujii *et al.* (1986) found that the low quality of sago starch is not only due to low level of processing techniques but also to other factors such as the freshness, maturity of raw materials (sago logs), storage of sago logs after harvesting, iron tools in starch production and the use of high-grade water for starch processing (Fujii *et al.*, 1986).

Consequently, other problems were also highlighted in recent years. With the above predicament in mind, this study was formulated to look into the basic characteristics of sago palm in terms of study of the physico-chemical properties of sago starch extracted from palms at different growth stages of maturity during the harvestable period.

#### 1.2 Specific Objectives

The specific objectives of this project were:

- (1) to determine the starch content of sago palm at different (maturity/harvestable) growth stages,
- (2) to characterise the physico-chemical properties (swelling factor, granule size and distribution, rheology, pasting and thermal profiles) of starch granules at different (maturity/harvestable) growth stages,
- (3) to study the composition of associated components such as soluble carbohydrates, phenolic compounds, lignin and non-starch polysaccharides, in the sago pith at different (maturity/harvestable) growth stages,

#### 1.3 Research protocol

This study commenced with the identification of sago palms of different maturity. These palms were chopped and sections of the trunk were brought back to the laboratory where the core pith samples were extracted using the borer tool. The remaining pith was chopped into smaller pieces and blended where sago starch was extracted manually from the blended pith. The dried sago flour was used for determination of physico-chemical properties whereas the core pith samples were used for determination of the composition of associated components in the pith.

The results collected will be analysed using ANOVA to find out whether there is any difference in the properties between the stages. The final conclusion can be made after considering the different physico-chemical properties of sago starch and the composition of associated components in the pith at the different (maturity) growth stages.

#### 2 LITERATURE REVIEW

#### 2.1 The Sago Palm

#### 2.1.1 Taxonomy

Sago palm belongs to the orders *Arecales* Nakai (Heywood, 1993), family *Palmae* Jussieu, subfamily *Calamoideae* Griffith, tribe *Calameae* Drude, subtribe *Metroxylinae* Blume and genus *Metroxylon* Rottboell (Uhl and Dransfield 1987). *Metroxylon* has previously been classified in the subfamily *Lepidocaroideae* (Moore, 1973) but this name has been changed back to *Calamoideae* by Uhl and Dransfield (1987), in agreement with the original classification of Griffith (1844). Further to this, Beccari (1918) and Rauwerdink (1986) also attempted to classify and distinguished the species of *Metroxylon* based on the fruit morphology and size among other considerations in species and subspecific classifications (Jong, 1995). Nevertheless, the two more important starch-producing species in the Malaysia and Indonesia regions are *Metroxylon sagu* Rottb. and *Metroxylon rumphii* Mart., of which the latter has spines on the petioles, spathes and even the leaflets (Sastrapradja and Mogea, 1977; Flach, 1984).

#### 2.1.2 Historical Origin and Distribution

The *Metroxylon*, the most widely known and exploited for food, has a distribution ranging from the Santa Cruz islands in the east to South Thailand in the west, from the Kai-Aru islands in the south to Mindanao in the north. The most dense distribution of *Metroxylon* appears to be the Moluccas, with Ceram (Seran) as centre and New Guinea (AVÉ, 1977).

The *Metroxylon sagu* occurs naturally from the South Pacific islands, extending westward through Melanesia into Indonesia, Malaysia and Thailand, where cultivated plants are largely indistinguishable from wild species. In nature, the palms occur in clumps and in relatively pure stands, and occupy lowland freshwater swamps (Johnson, 1977).

The true sago palm (*Metroxylon sago* Rottb.) is one of the potential under-utilized food palm and grows well in tropical rain forest of Southeast Asia between 10° of northern and southern latitudes (Mathur *et al.*, 1998). In Sarawak, *Metroxylon sagu* is the only crop which flourishes in the low-lying swampy plain. For many centuries, the people inhabiting these swamp forests of Oya, Mukah, Igan, Balingian and Dalat districts, most of whom are Melanau, have lived off the palm (Morris, 1977). Since then, the total area of growth by sago palm in Sarawak by 1990 was recorded to be 19,720 hectares (Tie *et al.*, 1991) with distribution as tabulated in **Table 2.1**. The main areas are in Oya-Dalat and Mukah involving 32.6 and 28.0 % of the total areas.

**Table 2.1** Distribution of sago palm areas (Tie *et al.*, 1991)

Division	Location	Area (ha)	%
Kuching	-	*	_
Samarahan	-	*	-
Sri Aman	Pusa-Saratok	3,240	16.4
Sibu	Oya-Dalat	6,410	32.6
	Mukah	5,520	28.0
	Balingian	950	4.8
	Igan	1,570	8.0
Sarikei	Matu-Daro	570	2.0
	Maradong	740	3.7
Bintulu	Bintulu-Tatau	340	1.7
Miri	-	*	-
Limbang	Limbang	380	1.9
Kapit	-	*	-
	Total	19,720	100.0

<sup>\*</sup> Negligible acreage; very scattered patches too small to be mapped at 1:250,000 scale

In 1992, enhanced by the concerted effort from the state government, the total area planted with sago palm in Sarawak increased with the establishment of estate plantations by the Land Custody and Development Authority (or Lembaga Pembangunan dan Lindungan Tanah, PELITA) of Sarawak. The distribution of estate areas is tabulated in **Table 2.2**.

**Table 2.2** PELITA Sago Estate Development (PELITA, 2003)

	Mukah Plantation Dalat Plantation					- Grand		
Area (ha)	Phase I	Phase II	Sebakong	Sub- total	Phase I		Sub- total	total
Gross	1,852	5,472	7,320	14,644	1,729	4,169	5,898	20,542
Planted	1,800	6,000	6,087	13,887	1,600	3,834	5,434	19,321
Rehabilitated	0	2,374	0	2,374	0	0	0	2,374

#### **2.1.3** Botany

Starch accumulation in palms on a massive scale as found in *Metroxylon* is almost always associated with the hapaxanthic flowering method where starch is accumulated in the pith of the stem and is mobilized at the onset of the production of a mass of inflorescences in the axils of the most distal leaves, giving a "terminal" inflorescence state. As flowering proceeds, the stem apex aborts and flowering and fruiting are followed by the death of the stem (Dransfield, 1977). Due to the massive size and lengthy vegetative phase, vast quantities of sago are stored in the stems.

Commonly called 'rumbia' in Malay-speaking regions, the sago palm produces an erect trunk about 10 m tall and 75 cm thick, which bears a crown of large pinnate leaves 5 m long with short petioles and leaf bases which clasp the stem (Kiew, 1977).

The sago palm is soboliferous (suckering) and has a massive rhizome which produces suckers freely. Sago may be propagated from suckers or seedlings. The plant forms a rosette of leaves in the early stage. Trunk formation starts during the 3<sup>rd</sup> to 4<sup>th</sup> year growth of the palm (Kueh, 1977). Sago trunks may reach 7 to 15 m in length and attain an average girth of 120 cm at the base of the palm (Flach and Schuiling, 1989). The vegetative phase in the sago palm lasts 7 - 15 years during which time, excess photosynthate from the leaves is transported to the trunk and stored as starch. The pith is saturated with starch from the base of the stem upwards (Kraalingen, 1986) and at maturity, the trunk is fully saturated with starch almost to the crown (Lim, 1991a). At maturity an enormous, branched inflorescence develops at the top of the trunk which developed into primary axis dividing into secondary and tertiary axes. Flowering followed by fruiting occurred on the tertiary axes (Flach, 1977). After the mature fruits fall off, the palm will soon die (Kueh et al., 1987). The development of the inflorescence to the production of ripe fruits lasts about 2 years during which time the remaining leaves fall and the carbohydrate supply in the stem is exhausted (Kiew, 1977). A pictorial presentation of the life cycle of a sago palm is as shown in **Figure 2.1**.

In Sarawak, *Metroxylon sagu* is the preferred sago palm to be planted by the local farmers as the smooth sheathed and thornless nature of the palm makes it easier to manage. The criteria by which sago palms are selected for harvesting are poorly documented. The starch reserves are apparently at their maximum just before flowering and fruiting deplete these reserves, but scientifically little more is known of the timing of starch build-up (Ruddle *et al.*, 1978). In Indonesia and Sarawak, the general belief is that the felling of the sago palm is best carried out after flowering but before the fruiting stage (Tan, 1982; Lim, 1991a).

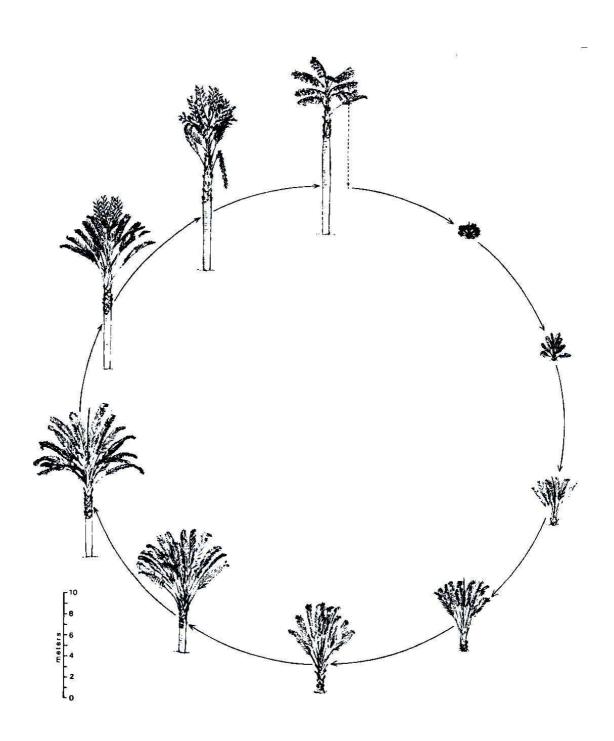


Figure 2.1 Life cycle of a sago palm (Schuiling and Flach,1985)

In Sarawak, the local farmers have classified the mature sago palms into the following five stages (**Table 2.3**). **Figures 2.2** to **2.6** showed the nature of sago palms at those growth stages.

**Table 2.3** Different physiological growth stages of sago palm (Lim, 1991a).

Growth stage	Palm description
Plawei	Palms that have reached maximum vegetative growth
Plawei Manit	Inflorescence emerging
Bubul	Inflorescence developing
Angau Muda	Flowering
Angau Tua	Fruiting

Lim (1991a) reported that the maximum starch yield per trunk is at the 'Angau Muda' stage (i.e. flowering stage) and declining at the 'Angau Tua' stage. No significant difference in starch yield among 'Plawei Manit', 'Bubul', and 'Angau Muda' stages was observed. Hence, Lim (1991a) concluded that the earliest stage at which a palm can be felled for maximum yield would be at 'Plawei Manit' stage.

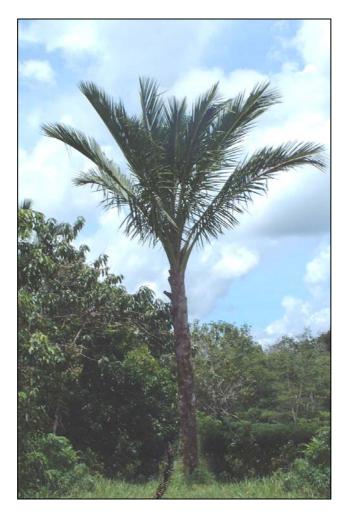
Jong (1995) gave a more comprehensive classification of growth stages as tabulated in **Table 2.4** based on his work with the farmers in Dalat, Sarawak. Jong (1995) found that the starch content is low in the early stages of trunk development and is mainly confined to the lower portion of the trunk. From the full trunk development stage onward, the pith is filled with maximum mean starch content of 22 % and density of 0.17 gcm<sup>-3</sup>. The high content and density of starch remain rather constant throughout the whole length of the trunk until the flowering stage. Thereafter, the level of starch decreases sharply, with a more pronounced dip at the topmost and bottommost positions of the trunk. This has been interpreted as the mobilisation of most of the starch for fruit development (Jong, 1995).

**Table 2.4** Stages of growth and development of sago palm in Sarawak (Jong, 1995)

Stage	Estimated age from planting (yr)	Local name	Duration of trunk growth (yr)	Growth description
1	1 – 5.5	Sulur	0	Rosette stage to the start of trunk formation; sucker-like young palm without visible trunk.
2	5.5	Angkat punggung	0	Starting of trunk formation; A transition between rosette and trunk growth. Short trunk are found upon removal of dead sheaths at the base of the palm at ground level.
3	7	Upong muda	1.5	Young trunk growth; trunks are about $1-2$ m in length.
4	8	Upong tua	2.5	25 % trunk growth; trunks are about $2-5$ m in length.
5	9	Bibang	3.5	50 % trunk growth; trunks are about $4-7$ m in length.
6	10	Pelawai	4.5	75 % trunk growth; trunks are about $6 - 8$ m in length.
7	11.5	Pelawai manit	6	Full trunk growth; full growth of harvestable trunk $(7-14 \text{ m})$ . Leaves become erect and small at the palm terminal. Appearance of whitish coloration on the stalks of these fronds.
8	12	Bubul	6.5	Bolting; Appearance of torpedo shaped flowering structures at the palm terminal. It is characterised by the elongation of the trunk at the top of the crown and frond reduction to bract-like structures.
9	12.5	Angau muda	7	Flowering; well-developed flowering structure with primary, secondary and tertiary flowering axes spreading out at the terminal. Flowers are in the pre- or post-anthesis stage.
10	13	Angau muda (same as stage 9)	7.5	Young fruiting; Fruits are about $20 - 30$ mm in diameter. Endosperms of the seeds (if any) are still soft and small. Most fronds are still intact and presumably functional.
11	14	Angau tua	8.5	Mature fruiting; Fruits are mature, of diameter 30 – 40 mm. Seeds (if any) are well developed with dark brown seed coat and bony endosperms. Most fronds are in senescent stage.
12	14.5	Mugun	9	Dying stage; most fruits have been shed and all fronds are in senescent stage.



**Figure 2.2** Plawei – palm that has reached maximum vegetative growth (photo copyright of CRAUN Research Sdn. Bhd.)



**Figure 2.3** Plawei Manit – inflorescence emerging palm (photo copyright of CRAUN Research Sdn. Bhd.)



**Figure 2.4** Bubul – inflorescence developing palm (photo copyright of CRAUN Research Sdn. Bhd.)



**Figure 2.5** Angau Muda – flowering palm (photo copyright of CRAUN Research Sdn. Bhd.)



**Figure 2.6** Angau Tua – fruiting palm (photo copyright of CRAUN Research Sdn. Bhd.)

#### 2.1.4 Extraction of Sago Starch

#### 2.1.4.1 Traditional Method of Extraction

The traditional method of extraction of sago starch can be classified into two levels, namely, the domestic level and the small-scale processing plants level (Flach, 1984).

The domestic level is practiced by the individual farmers where sago palms are felled and processed in the garden, thus without the need to transport the heavy trunk. After felling the trunk with an axe, it is split lengthwise. The pith is rasped by means of a chopper (Rhoads, 1977) or a small hoe (Höpfner, 1977), made from bamboo. The rasped mixture of fibre and pith is put on the wide end of a leaf sheath of the sago palm where a sieve is placed at its lowest end. Water is added to the mixture and then it is kneaded by hand. The fibres remain on top of the sieve while the water carrying the starch granules in suspension goes through the sieve and is caught in an old dugout canoe. The starch settles on the bottom and the excess water flows over the sides. After kneading, the fibrous remnants are discarded and the wet starch is taken out of the canoe (Flach, 1983; Ruddle *et al.*, 1978)

In the small-scale processing plant, sago trunks were cut into shorter length of 1 – 1.2 m and tied into raft and transported to the plant via rivers or man-made water system. Rasping is done using a board with nails in it. Some plants used an engine-powered rasps with which the pith is dug out of the split trunk and rasped. The rasped pith is trampled by foot on a platform. In some plants, rotating mesh washer made of metal or wood, or screen washers were used to separate the starch and coarse fibre. The starch slurry is channeled to a small settling ponds made of boards. Finally, drying of wet starch is done mostly in the sun (Flach, 1984; Ruddle *et al.*, 1978; Cecil, 2002; Oates & Hicks, 2002). Some small cottage mills produced only lamentak (wet

processed sago starch) or second grade quality flour which is sundried and unsieved wet sago starch (Zulpilip *et al.*, 1991).

#### 2.1.4.2 Modern Method of Extraction

Currently, there are nine sago factories operating in Sarawak, seven of which are in Mukah-Dalat areas, and the rest in Igan-Sibu areas (Manan *et al.*, 2003). The modern method of extraction involved some modification to that of the small processing plant where new technologies for extracting starch are adopted by the large-scale factories. These factories are fully mechanized and the level of technology is mostly found in Sarawak.

The 30 cm log sections from the storage pond are first split lengthwise into about 8 segments. These segments are fed into slicers that slice the pith from the bark (Oates & Hicks, 2002). In certain other factories, the bark was first removed from sections of the logs. Each of the debarked sections of about 80 – 100 cm long, is fed into the mechanical rasper (with chrome nails mounted on one face of a disc or a drum). This rasped the pith into finer pieces which are fed into the hammer mill via conveyor belt (Manan *et al.*, 2003). The resulting starch slurry is made to pass through a series of centrifugal sieves to separate the coarse fibres. Further purification is achieved by separation in a nozzle separator through sieve bends. A series of cyclone separators have also been used to obtain very pure starch. Dewatering of starch is carried out using a rotary vacuum drum drier followed by hot air drying (Azudin & Lim, 1991).

#### 2.1.5 Quality of Sago Starch

The Malaysian Standards for sago flour was determined by the Standards and Industrial Research Institute of Malaysia (SIRIM). The two Malaysian Standards are MS468 for industrial sago starch and MS470 for edible sago starch.

#### 2.1.5.1 Industrial Grade Sago Starch

In 1976, the Malaysian Standard MS468 Specification for Industrial Sago Starch defined industrial sago starch as "the processed starch obtained from the sago palm (*Metroxylon sagu*, *Metroxylon rumphii*) for use in the textile industry, as well as for manufacture of glucose, dextrines, monosodium glutamate, industrial alcohol and other industries requiring further conversion of sago starch." The requirements for industrial sago starch are tabulated in **Table 2.5**.

**Table 2.5** Requirements for Industrial Sago Starch (MS468, 1976)

No.	Characteristics	Requirements
1.	Starch content	60.0 % minimum
2.	Moisture content	15 % maximum
3.	Total ash (dry basis)	0.5 % maximum
4.	Crude Fibre (dry basis)	1.0 % maximum
5.	Particle size (thru' sieve of mesh 125)	65 % minimum
6.	Colour (tintometer readings)	0.4  red + 0.5  yellow
7.	pH of aqueous extract	4.0 minimum

#### 2.1.5.2 Edible Grade Sago Starch

As the sago industries grew over the years, the commercial factories also improved in the production of sago starch. In 1992, the first revision of Malaysian Standard MS470 Specification for Edible Sago Starch was initiated. Edible sago starch

was defined as starch in the form of fine powder derived from the trunk of the sago palm through the process of extraction and purification. The requirements for Edible Sago Starch are tabulated in **Table 2.6**.

**Table 2.6** Requirements for Edible Sago Starch (MS470, 1992)

No.	Characteristics	Requirements
1.	Moisture content	13 % maximum
2.	Total ash (dry basis)	0.2 % maximum
3.	pH of aqueous extract	4.5 - 6.5
4.	Crude fibre (dry basis)	0.1 % maximum
5.	Peak viscosity (6 % dry basis suspension)	600 AU
6.	Colour ("L" value)	90 minimum
7.	Sulphur dioxide	30 ppm maximum
8.	Particle size (thru' 125 µm or 120 mesh size)	99 % minimum

#### 2.1.6 Utilisation of Sago Starch

#### 2.1.6.1 Traditional Uses

Sago flour has been used as a staple food by the Melanaus in the Third Division of Sarawak (Sim, 1986). It is widely used for making keropok (Shrimp crackers) (Sidaway & Balasingam, 1971; Ong, 1979). Various food recipes using sago flour are known, e.g. *limut rampai*, sago dumplings in egg gravy, sago *choy suey*, sago curry, sago-stuffed *suntong*, savoury sago pancakes, sago with coconut milk, sago broth, sago hot pot, sago pudding, sago cones and *tabaloi* biscuits (Anon, 1980). Sago flour is also used in jellies, puddings and soups as sago pearls (Akiyama, 1966; Takahashi, 1986; Bujang & Ahmad, 2000)

#### 2.1.6.2 Uses in Food Industries

Sago flour is also used in some small-scale industries in Sarawak (**Table 2.7**). Due to its viscous property upon gelatinization, starch has potential to be used as thickener in the production of soup and baby food as well as additives in food products (Chulavatnatol, 2002; Zulpilip *et al.*, 1991; Takahashi, 1986; Ngudiwaluyo *et al.*, 1998).

**Table 2.7** Uses of sago flour in small industries (Sim, 1986).

Types of industries	Remarks on the use of sago flour
Noodles	25 % incorporation may cause slight changes in colour. Less fresh looking.
Chilli and tomato sauce	20 -30 % sago flour in the sauce is acceptable but reported to be less viscous
Biscuits	Moisture < 5 % of flour is necessary
Chips	Product is acceptable
Kway teoh (flat noodles)	20 % sago flour will make <i>kway teoh</i> harder and darker and no difference in taste is noted
Bread	25 % sago flour is incorporated and no difference in taste, texture or colour is noted
Buns	20 % sago flour is acceptable

#### 2.1.6.3 Uses in Non-Food Industries

The potential of sago starch to be used in the non-food industries was also exploited such as in the making of biodegradable plastic (Griffin, 1977; Pranamuda *et al.*, 1998; Odusanya *et al.*, 2000), as extender in urea formaldehyde adhesives (Solichin, 1986; Sumadiwangsa, 1985), as a finishing agent in the industrial production of paper and for sizing in textile industry. It is also a component of glue for sticking the sheet together in plywood manufacturing industry as well as for making glue gel and

liquid glue in paper box industry and offices (Chulavatnatol, 2002; Bujang & Ahmad, 2000) and in the manufacture of adhesives (Zulpilip *et al.*, 1991).

#### 2.1.6.4 Uses in Biotechnology

Like other starches, sago starch is also used in the production of ethanol and alcohol gasahol (Pranamuda *et al.*, 1995; Ishizaki & Tripetchkul, 1995; Haska, 1995), sugar metabolism and hydrolytic product (Hisajima *et al.*, 1995; Zulpilip *et al.*, 1991) and monosodium glutamate (Zulpilip *et al.*, 1991), as a substrate in the fermentation of acetone-butanol-ethanol (Gumbira *et al.*, 1996), and in the production of cyclodextrin (Solichien, 1995) and lactate industry (Ishizaki, 2002).

#### 2.1.6.5 Other Industries

Sago starch is also used in the animal & poultry feed formulation (Lim, 1991b). In the form of modified starches, sago starch is used as filler in pharmaceutical industry, as a replacement for hydroxymethyl cellulose to control fluid loss in the petroleum industries (Issham *et al.*, 1995; Bujang & Ahmad, 2000). To further enhance the utilization of sago starch, modification such as cross-linking and hydroxypropylation process was carried out on the sago starch. These processes can alter the properties of sago starch such that the applications in food industries are extended as thickeners, stabilizers and texturisers (Haryadi & Kuswanto, 1998; Bujang & Ahmad, 2000). Other researchers carried out physical modification to obtain pregelatinised and cold water soluble starches.

#### 2.2 Starch

Starch is the most widely produced carbohydrate by plants and is the major reserve polysaccharides of green plants (Morrison and Karkalas, 1990). Starch granules synthesized in amyloplasts and is deposited in the form of tiny granules in the major depots of seeds, tubers and roots. It is a source of energy and carbon for the developing plant, and it is the principal food of many animals, including man.

Starch granule is not chemically homogenous and can be separated into the simpler component amylose, a mixture of essentially linear molecules, and amylopectin, a mixture of highly branched polymers (Greenwood, 1976). In starch granules, the amylose and amylopectin molecules are radially oriented with their single reducing end-groups towards the centre or hilum, and synthesis is by apposition at the outer non-reducing ends (Nikuni, 1978; Blanshard, 1979; French, 1984).

#### 2.2.1 Amylose and Amylopectin

Amylose is defined as a linear molecule of  $(1\rightarrow 4)$  linked  $\alpha$ -D-glycopyranosyl units (**Figure 2.7**), with some molecules slightly branched by  $(1\rightarrow 6)$ - $\alpha$ -linkages (French, 1984). In some species, amylose has a few phosphate groups, probably at C-6 of glucose residues. Depending on source, amylose has an average of 2 – 11 branch points and therefore 3 – 12 non-reducing chain ends per reducing end (Morrison & Karkalas, 1990). Amylose is found with molecular weights ranging from  $10^5 - 10^6$  and with the number of glucose residues per molecule, (DP) ranging from 500 to 5000 (Galliard & Bowler, 1987).

Amylopectin is a branched polysaccharides composed of hundreds of short  $(1\rightarrow 4)$ - $\alpha$ -glucan chains, which are interlinked by  $(1\rightarrow 6)$ - $\alpha$ -linkages (**Figure 2.7**).

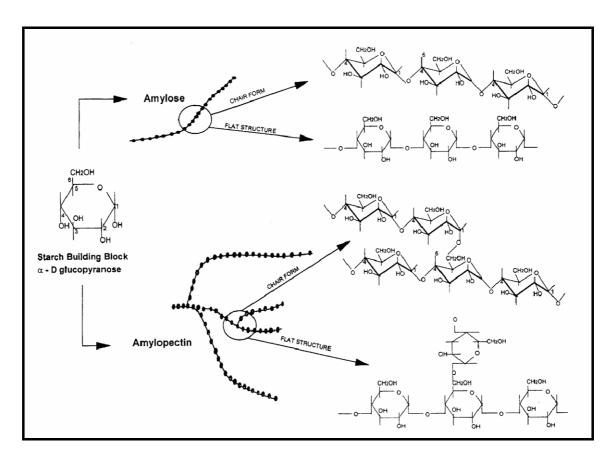


Figure 2.7 Linear and branched starch polymers (Murphy, 2000)