



UNIVERSITI PUTRA MALAYSIA

MATERNAL TOXICITY AND TERATOGENIC EFFECTS OF *Jatropha curcas* L. OIL IN PREGNANT *Sprague dawley* RATS

YON THANNIA BINTI SAMAT

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**MASTER OF SCIENCE
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By

YON THANNIA BINTI SAMAT

**Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in
Fulfillment on the Requirements for the Degree of Master of Science**

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment
of the requirement for the degree of Master of Science

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October 2012

Chair: Associate Professor Sabrina Sukardi, PhD

Faculty: Medicine and Health Sciences

Jatropha curcas (L.) (*Euphorbiaceae*), is a large shrub, attaining 3-4 m in height, common in Brazil and also found in India and Africa in semi-wild conditions. Laboratory studies on the effects of *Jatropha curcas* on pregnancy is limited and the number of experimental animals used in most studies were insufficient for any firm conclusions to be drawn. Research however has shown that it has pregnancy terminating effects in rats and mice and was used widely in some African countries for contraceptive intentions. Feeding studies also showed that the whole seeds were highly toxic. Curcin, a toxic protein isolated from the seeds, inhibits protein synthesis in *in vitro* studies even though it is less toxic than ricin and abrin. The oil of *Jatropha* contains irritant phorbol esters which causes purgative and skin irritant effects and possess tumour-promoting (co-carcinogenic) properties. In this study, pregnant Sprague-Dawley rats were administered *Jatropha* crude oil (JCO) orally at doses of 0.175, 0.35 or 0.7 g/ml during embryogenesis (Gestation day (GD) 1-7) for early gestation group and during organogenesis on GD 8-14 for late gestation group to

examine any toxic effects. On day 21st of pregnancy, the rats were anesthetized with chloroform. Ovaries and uteri were removed by Caesarean section. Heart, ovaries, placenta, liver, intestines, stomach, kidneys, and lungs of dams were collected and weighed. In addition, number of fetuses were recorded and examined for obvious external malformations before subjected to fetal staining to assess teratogenic effects. Placentas were stored in 10% buffered formalin for subsequent histopathology examination to observe effects of JCO on placental morphology. Results were reported as means \pm S.E.M. Data were analyzed with SPSS. Two-way ANOVA followed by Duncan post hoc test were used to determine the degree of significance for the various mean variables obtained and $p < 0.05$ was considered significant. Body weight of rats exposed to JCO at doses 0.35 and 0.7 g/ml examined were significantly lower ($p < 0.05$) than those of controls who only received corn oil. No fetuses were observed with external malformations in this study but for skeletal malformations, variations or abnormalities observed in fetuses from treated groups include dumbbell shape vertebrae, split vertebrae, wavy ribs, poorly ossified sternum and xiphisternum and hypoplastic sternum. Placentas of rats exposed to JCO showed histological changes in maternal-fetal interface, trophoblastic giant cell layers, and labyrinth layer with an increase in abnormal trophoblastic giant cells which has atypical shape with pyknotic and irregular nuclei. The results of this study indicate that JCO causes acute toxicity to the dams, induce teratogenic effects on fetuses and stimulate deleterious effects on placenta.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

**KESAN TOKSIK TERHADAP IBU DAN TERATOGENIK OLEH MINYAK
ASLI *Jatropha curcas* DALAM TIKUS *Sprague dawley* BUNTING**

Oleh

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Jatropha curcas (L.) (*Euphorbiaceae*), adalah sejenis pokok tidak berkayu dengan ketinggian 3-4 m, yang berasal dari Brazil dan juga India dan Africa dalam keadaan separa liar. Ujikaji makmal akan kesan *Jatropha curcas* semasa kebuntingan terhadap bilangan haiwan kajian yang digunakan dalam ujikaji kebanyakan juga tidak mencukupi untuk membuat sebarang kesimpulan yang kukuh. Penyelidikan telah menunjukkan bahawa ia mempunyai kesan keguguran pada tikus dan mencit dan juga digunakan dengan meluas di negara tertentu seperti Afrika untuk tujuan mencegah kehamilan. Kajian pemakanan telah menunjukkan bahawa keseluruhan biji adalah sangat toksik. Curcin, sejenis protein bersifat toksik dari biji, boleh merencat sintesis protein dalam kajian *in vitro* walaupun ianya kurang toksik daripada ricin dan abrin. Minyak *Jatropha* mengandungi phorbol ester yang mengakibatkan cirit birit serta kegatalan kulit dan juga mengandungi bahan-bahan penyebab tumor. Di dalam kajian ini, tikus *Sprague-Dawley* yang bunting diberi minyak mentah *Jatropha* melalui mulut pada dos 0.175, 0.35 dan 0.7 g/ml ketika pembentukan embrio iaitu pada hari ke 1-7 kebuntingan bagi kumpulan awal

kebuntingan dan ketika pembentukan organ pada hari ke 8-14 kebuntingan bagi kumpulan akhir kebuntingan untuk mengkaji kesan toksiknya dalam tikus bunting. Pada hari kebuntingan yang ke 21, ibu tikus dikorbankan dengan overdos klorofom. Rahim serta ovari dikeluarkan secara kaedah potongan "Caesarean". Jantung, ovari, uri, hati, usus, perut, buah pinggang dan peparu ibu tikus dikeluarkan dan ditimbang. Bilangan janin dicatat dan kesemua janin diperhatikan untuk kecacatan luaran sebelum menjalani proses pewarnaan janin bagi mengkaji kesan teratogenik. Uri disimpan di dalam 10% cecair formalin bagi menjalani pemeriksaan histopatologi untuk mengkaji kesan minyak mentah *Jatropha* terhadap histologi uri. Keputusan dicatat dalam bentuk nilai min \pm SEM. Data dianalisis menggunakan ANOVA dua hala diikuti dengan ujian Duncan post hoc dan nilai $p < 0.05$ dianggap signifikan. Berat badan tikus yang terdedah kepada minyak mentah *Jatropha* dengan dos 0.35 dan 0.7 g/ml didapati lebih rendah secara signifikan ($p < 0.05$) berbanding berat badan tikus kawalan yang hanya menerima minyak jagung. Tiada janin yang mengalami kecacatan luaran dalam kajian ini tetapi untuk kecacatan tulang, variasi atau ketidak normalan yang dapat dilihat pada janin daripada kumpulan rawatan termasuklah vertebra berbentuk "dumbbell", vertebra terpisah, rangka berombak, sternum, dan xiphisternum yang kurang osifikasi serta sternum hipoplastik. Uri tikus dari kumpulan rawatan menunjukkan perubahan histologi pada interfasa ibu-janin, lapisan sel gergasi trophoblastik dan lapisan labirin dengan peningkatan dalam sel gergasi trophoblastik mempunyai bentuk atipikal dan nukleus yang piknotik dan berbentuk tidak sekata. Keputusan kajian ini menunjukkan bahawa minyak mentah *Jatropha* mengakibatkan toksisiti akut kepada ibu tikus, menyebabkan kesan teratogenik terhadap janin dan mengakibatkan kesan kerosakan pada uri.

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This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Master of Science. The members of the Supervisory Committee were as follows:

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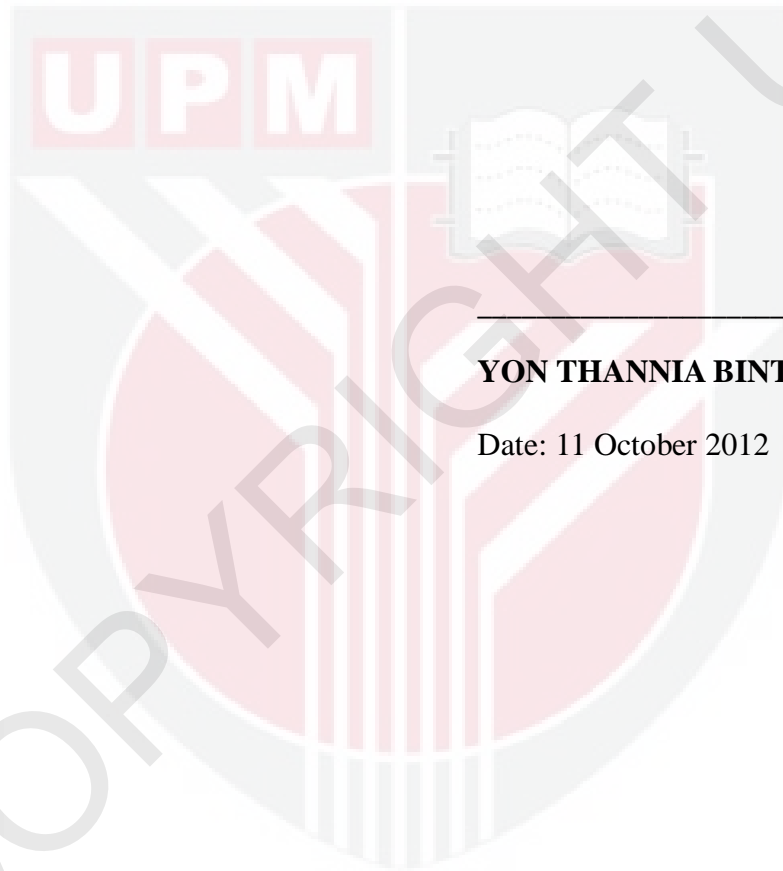
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DECLARATION

I declare that the thesis is my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously, and is not concurrently, submitted for any other degree at Universiti Putra Malaysia or at any other institution.



YON THANNIA BINTI SAMAT

Date: 11 October 2012

TABLE OF CONTENTS

	Page
ABSTRACT	ii
ABSTRAK	iv
ACKNOWLEDGEMENT	vi
APPROVAL	vii
DECLARATION	ix
LIST OF TABLES	xiv
LIST OF FIGURES	xvii
LIST OF ABBREVIATIONS	xx
CHAPTER	
1 INTRODUCTION	1
1.1 Background of Study	1
1.2 Problem Statement	3
1.3 Objectives	4
1.4 Hypothesis	5
2 LITERATURE REVIEW	6
2.1 Teratogens from plants	6
2.2 Distribution of <i>Jatropha curcas L.</i>	9
2.3 Botanical description of <i>Jatropha curcas L.</i>	10
2.4 Uses of <i>Jatropha curcas L.</i>	12
2.4.1 Oil Crop	12
2.4.2 <i>Jatropha curcas</i> as Folk Medicine	13
2.4.2.1 Latex	13
2.4.2.2 Leaves	14
2.4.2.3 Roots	14
2.4.2.4 Seeds	14
2.4.2.5 Fruits	15
2.4.3 Soil Enrichment	16
2.4.4 Feed	16
2.5 Toxic effects of <i>Jatropha curcas L.</i>	17
2.5.1 Toxic effects of phorbol esters of <i>Jatropha curcas L.</i>	19
2.5.2 Toxic effects of curcin of <i>Jatropha curcas L.</i>	20
2.6 Systemic description of clinical effects of <i>Jatropha curcas L.</i> toxicities	21
2.6.1 Cardiovascular	21
2.6.2 Respiratory	21
2.6.3 Neurological	21
2.6.3.1 Central Nervous System (CNS)	21
2.6.3.2 Autonomic Nervous System	21
2.6.3.3 Skeletal and Smooth Muscle	22
2.6.4 Gastrointestinal	22
2.6.5 Hepatic	22
2.6.6 Urinary	22
2.6.7 Endocrine and Reproductive Systems	22

2.6.8	Dermatological	23
2.6.9	Hematological	23
2.6.10	Allergic Reactions	24
2.7	Retinyl Palmitate	24
2.7.1	Overview of Retinyl Palmitate	24
2.7.2	Uses of Retinyl Palmitate	25
2.7.3	Research on Retinyl Palmitate	26
2.8	Maternal Toxicity	27
2.9	Rat Reproductive System	28
2.9.1	Female Reproductive Tract	29
2.9.2	Ovulation and Fertilization	30
2.9.3	Gestation Period	31
2.9.4	Trimester	31
2.10	Staging of Rat Development	33
2.10.1	Cleavage and Blastulae Stage (Day 1 to 5)	33
2.10.2	Gastrula Stage (Day 6 to 8.5)	34
2.10.3	Neurula Stage (Day 9 to 11)	35
2.10.4	Tail Bud Embryo Stage (Day 11.5 to 12.375)	36
2.10.5	Complete Embryo Stage (Day 12.5)	37
2.10.6	Metamorphosing Embryo Stage (Day 12.75 to 16)	37
2.10.7	Fetus Stage (Day 17 to 22)	38
2.11	Placenta	38
2.12	Rat Placenta Organization	39
2.13	Rat Trophoblast Cell Types of the Chorioallantoic Placenta	40
2.13.1	Cyto- and Syncytiotrophoblast	40
2.13.2	Giant Cells	41
2.13.3	Spongiotrophoblast	42
2.14	Decidua Tissue, Structure and Functions	42
2.14.1	Origin of Decidua	42
2.14.2	Decidual Cell Structure	44
2.14.2.1	Stromal Cells	44
2.14.2.2	Uterine Natural Killer (uNK) Cells	44
2.14.2.3	Decidual Macrophages	45
2.14.2.4	Dendritic (Antigen Presenting) Cells	45
2.14.3	Functions of the Decidual Cells	45
2.14.3.1	Isolation of the Implanting Blastocyst	45
2.14.3.2	Nutrient Provision	46
2.14.3.3	Hormone Production	46
2.14.3.4	Gap Junctions	46
2.15	Similarities between Rat and Human Placenta	47
2.16	Differences between Rat and Human Placenta	48
3	MATERIALS AND METHODS	51
3.1	Production of <i>Jatropha</i> crude oil	51
3.2	Preparation of Retinyl Palmitate (Positive Control)	52
3.3	Determination of JCO Doses	53
3.4	Animals	54
3.5	Mating Methods	54
3.6	Vaginal Smear Procedure	55
3.7	Experimental Procedure	56

3.8	Fetus Staining	58
3.9	Fetus Analysis	59
3.10	Maternal Toxicity Assessment	60
	3.10.1 Placenta, Kidney and Liver Slide Preparation	60
	3.10.2 Morphology of Kidney and Liver	61
3.11	Statistical Analysis	62
	3.11.1 Maternal, fetuses and organs weight	62
	3.11.2 Skeletal Analysis	62
	3.11.3 Histopathology of Placenta	63
4	RESULTS	64
4.1	Effects of <i>Jatropha curcas</i> crude oil (JCO) on maternal body weight	64
4.2	Effects of JCO on maternal organs weight	67
	4.2.1 Heart	67
	4.2.2 Lung	68
	4.2.3 Stomach	69
	4.2.4 Liver	70
	4.2.5 Kidneys	71
	4.2.6 Intestine	72
	4.2.7 Ovary	73
	4.2.8 Uterus	74
4.3	Effects of JCO on number of females with fetuses	76
4.4	Effects of JCO on fetuses	76
	4.4.1 Effects of JCO on number of fetuses	76
	4.4.2 Effects of JCO on placental weight	77
	4.4.3 Effects of JCO on fetus body weight	78
	4.4.4 Effects of JCO on fetal size	79
	4.4.4.1 Effects of JCO on fetal head size	79
	4.4.4.2 Effects of JCO on fetal tail size	80
	4.4.4.3 Effects of JCO on fetal body size	81
4.5	Examination on skeletal anomalies of fetuses	83
	4.5.1 Dumbbell Shape Vertebrae	83
	4.5.2 Split Vertebrae	85
	4.5.3 Wavy Ribs	86
	4.5.4 Ossification Centre at the Vertebrae	88
	4.5.5 Not Well Ossified Sternum	89
	4.5.6 Not Well Ossified Xiphisternum	90
	4.5.7 Dumbbell Shape at Sternum	92
	4.5.8 Split Sternum	94
	4.5.9 Hypoplastic Sternum	95
	4.5.10 Absence of Sternum	97
4.6	Examination for Placental Anomalies	98
	4.6.1 Examination for Placental Anomalies during Early Gestation	
	Treatment Group	98
	4.6.1.1 Placental Layers	98
	4.6.1.2 Trophoblastic Giant Cells	99
	4.6.2 Examination for Placental Anomalies during Late Gestation	
	Treatment group	101
	4.6.2.1 Placental Layers	101

4.6.2.2	Trophoblastic Giant Cells	102
5	DISCUSSION	106
5.1	Teratology	106
5.2	Toxic effects of <i>Jatropha</i> crude oil (JCO)	108
5.3	Assessment of Toxicity	111
5.3.1	Organs Weight	111
5.3.2	Effects of JCO on Number of Females with Fetuses	112
5.3.3	Effects of JCO on Fetus Body Weight	113
5.3.4	Skeletal Examination	114
5.3.5	Effects of JCO on Placenta	115
5.3.6	Effects of JCO on Liver and Kidneys	119
5.4	Retinyl Palmitate	120
5.4.1	Toxicity of Retinyl Palmitate	120
5.4.2	Metabolism of Retinyl Palmitate	122
6	CONCLUSIONS AND FUTURE RECOMMENDATIONS	124
	REFERENCES	127
	APPENDICES	141
	BIODATA OF STUDENT	151