



اَوْبُو سَيِّدِي تَيْكُونُو لُو كِي مَبَارَا  
UNIVERSITI  
TEKNOLOGI  
MARA

**DETECTION AND QUANTIFICATION OF ASCORBIC ACID IN  
YELLOW CRIMSON WATERMELON (*Citrullus lanatus*) BY  
USING REVERSE-PHASE HIGH PERFORMANCE LIQUID  
CHROMATOGRAPHY (HPLC)**

**By:**

**NURARIFAH BINTI ABD RAHIM**

Thesis Submitted in Partial Fulfilment of the Requirement for  
Bachelor of Medical Laboratory Technology (Hons),  
Faculty of Health Sciences, Universiti Teknologi MARA

2017

## **DECLARATION**

I hereby declare that this thesis is my original work and has not been submitted previously or currently for any other degree at UiTM or any other institutions.

Signature:

Name: Nurarifah Binti Abd Rahim

Matric Number: 2013230256

Date: 21<sup>st</sup> July 2017

## **ACKNOWLEDGEMENT**

In the name of Allah, the Most Gracious and the Most Merciful

Alhamdulillah, all praises to Allah for the bliss, strength, health and His mercy for me and my friends throughout the durations along this journey to complete our final year research project. First and foremost, I would like to express my gratitude towards my beloved parents and all my family members for their pray, help, supportive morally and physically and always been there for me the whole time in completing this project. Secondly, I would like to express my gratitude to my respected final year project supervisor Dr Wan Mazlina Bt Md Saad and Dr Fatimah Bt Salim for their continual support, guidance, supervision, advice and knowledge in completing this thesis. I would also dedicate my special thanks to all lecturers, all laboratory personnel and staff from Centre of Medical Laboratory Technology and Atta-ur-Rahman Institute for Natural Product Discovery (AuRIns) for their guidance, help and moral support in the making and completing this project. Special thanks to Faculty of Health Sciences, Department of Medical Laboratory Technology and Department of Postgraduate Study, UiTM Puncak Alam for funding my study and providing me with necessary laboratory facilities throughout the time of completing this study. This study will not be completed without the help from postgraduate student, Rasdin Ridwan that have been very helpful with his assistance and knowledge from the beginning of the study, a big thank you and may Allah bless you. Not forgetting my deep gratitude goes to my group members for their commitment, support, time, contribution, help and this memorable friendship during the whole time of completing this thesis. Last but not least, my sincere gratitude for those who have been directly and indirectly contributed in accomplishing this final year research project.

# TABLE OF CONTENTS

## Contents

Title Page	i
Declaration	ii
Intellectual Properties	iii
Acknowledgements	vi
Table of Contents	vii
List of Tables	ix
List of Figures	x
List of Abbreviations	xi
Abstract	xii
<b>CHAPTER 1</b>	
1.1 Overview .....	1
1.2 Problem statement .....	2
1.3 Research Objectives .....	3
1.3.1 General Objective.....	3
1.3.2 Specific Objective .....	3
<b>CHAPTER 2</b>	
2.1 Overview of watermelon .....	4
2.1.1 Characteristics of Watermelon.....	4
2.1.2 Classification of Watermelon.....	5
2.1.3 Pharmacology Properties .....	6
2.1.4 Chemical constituent.....	6
2.2 Ascorbic acid .....	8
2.2.1 Sources of Ascorbic acid.....	8
2.2.2 Chemical and physical characteristics of Ascorbic acid .....	8
2.2.3 Biosynthesis of Ascorbic acid.....	9
2.2.4 Role of Ascorbic acid.....	10
2.2.5 Ascorbic acid as antioxidant .....	11
2.2.6 Ascorbic acid in health and diseases .....	11
2.3 Thin Layer Chromatography (TLC).....	12
2.3.1 Thin Layer Chromatography .....	12
2.3.2 Principle of Thin Layer Chromatography .....	12

## ABSTRACT

### DETECTION AND QUANTIFICATION OF ASCORBIC ACID IN YELLOW CRIMSON WATERMELON (*Citrullus lanatus*) BY USING REVERSE-PHASE HIGH PERFORMANCE LIQUID CHROMATOGRAPHY (HPLC)

Watermelon or *Citrullus lanatus* (Thunb.) Matsum. And Nakai is a good source of phytonutrients and antioxidants such as Vitamin C, water soluble vitamins which is an essential micronutrient that have several important biological functions. Vitamin C as a potent antioxidant can eliminates reactive oxygen and nitrogen species and protective against oxidative damage. Due to lack of an enzyme human cannot synthesize ascorbic acid hence, it has to be supplemented through fruits such as watermelon. However, there are insufficient studies on ascorbic acid identification and quantification by using HPLC with better extraction method that can prevent ascorbic acid oxidation in local yellow watermelon. Therefore, this study was conducted to identify and quantify ascorbic acid compound in local yellow watermelon by using 3% Ortho-phosphoric and 8% Acetic acid extraction with 0.1% Ortho-phosphoric : Acetonitrile (95:5) mobile phase in reverse-phase High Performance Liquid Chromatography (HPLC). The retention time for the ascorbic acid in the chromatogram is 2.798 min for both standard and sample. Standard curves were linear over the concentration range from 100 to 500 µg/mL and the limits of detection and quantification were 0.47 and 1.43 µg/mL, respectively. The method was validated by doing spike recovery. The concentration of ascorbic acid in yellow flesh of local watermelon was found to be 19.05 µg/mL with percentage of 0.00002%. In conclusion, it can be stated that reverse-phase HPLC is the most preferred method to identify the presence of ascorbic acid. In addition, 3% Ortho-phosphoric and 8% Acetic acid in extraction method was a good method in order to prevent ascorbic acid oxidation.

**Keywords:** *Citrullus lanatus*, yellow watermelon, Ascorbic acid, HPLC, analytical method, Malaysia.