THE UNIVERSITY OF MANITOBA

PHENOLIC COMPOUNDS FOUND IN CANOLA:

CONVERSION TO LIGNAN

AND

EFFECTS ON GLOBULAR PROTEIN PROPERTIES

By

Maria I. Rubino

A Thesis
Submitted to the Faculty of Graduate Studies
in Partial Fulfillment of the requirements
for the degree of

DOCTOR OF PHILOSOPHY

Food and Nutritional Sciences
University of Manitoba
Winnipeg, Manitoba

October, 1995

National Library
of Canada

Acquisitions and
Bibliographic Services Branch

395 Wellington Street
Ottawa, Ontario
K1A 0N4

Bibliothèque nationale
du Canada

Direction des acquisitions et
des services bibliographiques

395, rue Wellington
Ottawa (Ontario)
K1A 0N4

Your file *Votre référence*

Our file *Notre référence*

ISBN 0-612-13489-X

Canada

PHENOLIC COMPOUNDS FOUND IN CANOLA: CONVERSION TO LIGNAN AND EFFECTS ON GLOBULAR PROTEIN PROPERTIES

BY

MARIA I. RUBINO

A Thesis submitted to the Faculty of Graduate Studies of the University of Manitoba in partial fulfillment of the requirements of the degree of

DOCTOR OF PHILOSOPHY

To my parents: Elbio, Aida, Rita and Frank.

ACKNOWLEDGEMENT

I would like to express my sincere thanks to my supervisor, Dr. Susan D. Arntfield for her invaluable advice, guidance, support, friendship and patient during the course of this research project. Special thanks are extended to Dr. James Charlton for his enthusiasm and help in identifying the thomasidioic acid. I wish to thank the members of my committee Dr. P. McVetty, Dr.B. Watts and Dr. W. Bushuk.

I would like to thanks the technical assistance of my friend Aniko Bertnatsky, specially for her work and patient during the HPLC analysis. Technical assistance, support and friendship was generously provided by Jim Roger. I would like to acknowledge the help, support and friendship of Malgorzata Scygiel and Georgina Mejias.

I am very grateful for the financial assistance I received from the Natural Science Research Council (NSERC) grant to Dr. Susan Arntfield.

During the course of this work I have had the opportunity to make a number of friends which make this time unforgettable, exciting and very enjoyable. In addition I would like to thank my family for their understanding and support.

Finally, I want to thank my Father Elbio whose enthusiasm, curiosity and faith in the future have been a constant source of inspiration.

ABSTRACT

Phenolic compounds in plant materials represent an area of concern, because products obtained from plant seeds containing phenolic compounds have low sensory characteristics and nutritional value. Sinapic acid and sinapine are the preponderant compounds found in canola, and knowledge of their behaviour is important if increased usage of canola protein is to be realized. The goals of this work were to elucidate the changes in the phenolic compounds under different conditions of pH and to examine how the phenolic compounds and their by-products bind to the protein and affect protein functionality.

Changes in phenolics were followed using U.V. spectroscopy, high performance liquid chromatography (HPLC) and nuclear magnetic resonance (NMR) spectroscopy. Binding of purified phenolic compounds and associated by-products to bovine serum albumin (BSA) and canola protein as a function of pH and salt concentration, was evaluated using equilibrium dialysis in conjunction with HPLC analysis. Gelation properties of proteins at similar levels of phenolics and by-products were evaluated with dynamic rheology.

It has been shown that sinapic acid readily converts to a lignan, thomasidioc acid, when exposed to alkaline aqueous buffer; this identification was confirmed by comparison with a synthesized authentic sample of thomasidioic acid. This conversion is complete at pH 8.5 but there is only partial conversion at pH 7. The reaction can be completely controlled

by the elimination of oxygen, or partially controlled by the inclusion of ascorbic acid.

It was found that sinapic acid and sinapine interact with the proteins mainly through electrostatic interaction, whereas the thomasidioic acid and sinapine by-products react through hydrophobic interactions. The ligand involved in binding was pH dependent. Furthermore, there was more binding to the canola protein. In general the presence of either ligand reduced the three dimensional network as well as the elasticity of the gel. This reduction was greatest when binding was greatest.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF APPENDICES