

Copyright is owned by the Author of the thesis. Permission is given for a copy to be downloaded by an individual for the purpose of research and private study only. The thesis may not be reproduced elsewhere without the permission of the Author.

STUDIES ON CAROTENOID  
METABOLISM

By

N.A. WORKER

Biochemistry Department  
Massey Agricultural College

.....

Being a Thesis Submitted to the University of New Zealand  
in Partial Fulfilment of the Requirements for The Degree  
of Doctor of Philosophy.

November 1957

## LIST OF CONTENTS

	Page.
<u>SECTION I</u>	
STUDIES ON THE UTILIZATION OF PARENTERALLY ADMINISTERED CAROTENOIDS.	
INTRODUCTION	1
EXPERIMENTAL MATERIALS AND METHODS	8
CHAPTER I. The effect of the thyroid on the conversion of intravenously administered aqueous dispersions of carotene to vitamin A in the rat.	18
CHAPTER II. Studies on the site of conversion of intravenously administered aqueous dispersions of carotene to vitamin A in the rat and rabbit	
I. The effect of complete hepatectomy and evisceration.	28
CHAPTER III. Further studies on the site of conversion of intravenously administered aqueous dispersions of carotene to vitamin A in the rat.	
II. The effect of removal of the lungs and decapitation.	40
CHAPTER IV. Studies on the <u>in vitro</u> conversion of carotene to vitamin A in tissues from the rat, guinea pig and sheep	47
CHAPTER V. A study of some factors affecting the utilization of parenterally administered aqueous dispersions of carotene to vitamin A.	78
CHAPTER VI. Further studies on the effect of tocopherol on the utilization of intravenously administered aqueous dispersions of carotene.	101
CHAPTER VII. The utilization of aqueous dispersions of carotene by guinea pigs and carotene and vitamin A by lactating goats.	116

	Page.
CHAPTER VIII. The utilization of intra- venously administered carotene and vitamin A emulsions by rats and goats.	130
CHAPTER IX. Integrating summary to Section I.	139
REFERENCES	144
 SECTION II	
A STUDY OF SOME FACTORS AFFECTING THE VITAMIN A POTENCY OF MILK FAT - With Particular Reference to Factors Affecting the Summer Decline in the Vitamin A Potency of New Zealand Milk Fat.	
INTRODUCTION	149
EXPERIMENTAL MATERIALS AND METHODS	165
CHAPTER X. A survey of the changes occurring in the levels of various blood plasma and milk fat constituents in cows grazing New Zealand pastures over the spring and summer.	176
CHAPTER XI. Studies on the utilization of carotene from summer pasture and from arachis oil by the cow.	192
CHAPTER XII. The influence of various nutritional factors on the utilization of carotene administered orally to rats.	204
CHAPTER XIII. Integrating summary to Section II.	217
REFERENCES	222



# LIST OF TABLES

Table		Page.
1.	The vitamin A content of the blood plasma and liver of partially deficient rats.	10
2.	Demonstrating the uniformity of vitamin A ester, relative to vitamin A alcohol, in the blood of partially deficient rats at various intervals after injection of 400 $\mu$ g of carotene in Tween.	13
3.	The effect of thyroid activity on the conversion of carotene to vitamin A 24 h after intravenous administration of carotene as an aqueous dispersion in Tween to rats partially deficient in vitamin A.	22
4.	The effect of thyroid activity on the conversion of carotene to vitamin A 24 h after intravenous administration of carotene as an aqueous dispersion in Tween to rats partially deficient in vitamin A.	23
5.	The effect of the thyroid on the remission of xerophthalmia and on weight increases in rats following intravenous injection of carotene as an aqueous dispersion in Tween.	24
6.	Appearance of carotene and vitamin A alcohol in the blood of partially deficient hepatectomized and hepatectomized-visceroctomized rats after intravenous injection of Tween dispersions of carotene.	34
7.	Appearance of carotene and vitamin A alcohol in the blood of hepatectomized rabbits after intravenous injection of Tween dispersions of carotene.	35
8.	Appearance of carotene and vitamin A in the blood and livers of pneumonectomized and decapitated rats 5 min after injection of carotene in Tween.	44
9.	Perfusion of the intact partially deficient rat with Tween dispersions of carotene and vitamin A alcohol in physiological saline for varying periods up to 4 h.	62
10.	Perfusion of isolated organs from partially deficient rats with Tween dispersions or emulsions of carotene in physiological saline for varying periods up to 4 h.	63
11.	Perfusion of isolated organs from partially deficient rats with Tween dispersions or emulsions of vitamin A alcohol in physiological saline for varying periods up to 4 h.	64

12.	Perfusion through the lumen of the intestine of the partially deficient rat both <u>in vivo</u> and <u>in vitro</u> with Tween dispersions of carotene, vitamin A alcohol and retinene in saline, or with emulsions of carotene or vitamin A alcohol in saline.	65.
13.	Incubation of tissue slices and tissue homogenates from partially deficient rats with carotene, vitamin A alcohol and retinene in Tween, and with carotene and vitamin A alcohol in emulsion form for periods of from 2 to 4 h.	66
14.	Incubation of whole and chopped rat intestines in physiological saline with carotene and vitamin A alcohol as emulsions and as Tween dispersions for various intervals up to 16 h.	67
15.	Incubation of tissue slices and tissue homogenates from normal guinea pigs with Tween dispersions and emulsions of carotene and vitamin A alcohol in physiological saline for varying periods up to 24 h.	68
16.	Incubation of blood from the rat, guinea pig and sheep with various preparations of carotene and vitamin A alcohol for periods up to 16 h.	69
17.	Incubation of blood, separately and in combination, with carotene and with intestinal homogenate, to demonstrate esterification of true vitamin A alcohol and its chromatographic separation from oxidation products of carotene which interfere with the antimony trichloride reaction.	70
18.	Utilization of aqueous carotene dispersions in Tween administered by various routes to partially deficient rats, killed 4 days after dosing.	87
19.	Effect of dose level, interval between dosing and slaughter, repeated dosing and vitamin A status on the utilization of aqueous carotene dispersions in Tween administered intravenously to partially deficient rats.	88
20.	Effect of xanthophyll, tocopherol and benzoyl peroxide on the utilization of aqueous carotene dispersions in Tween administered intravenously to rats, of mean weight 300 g, killed 24 h after dosing.	89
21.	Appearance of carotenoid pigments and vitamin A in the blood and liver of rats partially deficient in vitamin A after the intravenous injection of the pigments as Tween dispersions.	90

Table		Page.
22.	Levels of tocopherol in rat tissues at various time intervals after the intravenous injection of aqueous dispersions of $\alpha$ -tocopheryl acetate in Tween.	105
23.	The effect of preliminary, simultaneous or subsequent intravenous injections of an aqueous dispersion of $\alpha$ -tocopheryl acetate in Tween on the liver and blood levels of vitamin A formed from carotene similarly administered.	106
24.	Appearance of carotene and vitamin A in whole rat carcasses at various time intervals after the intravenous injection of an aqueous dispersion of carotene in Tween.	107
25.	Appearance of carotene and vitamin A in the blood and liver of young guinea pigs after intravenous administration of aqueous dispersions of carotene in Tween.	121
26.	Utilization of aqueous dispersions of carotene and vitamin A alcohol and palmitate injected intravenously into lactating goats.	122
27.	Utilization of aqueous dispersions of carotene and of vitamin A alcohol and palmitate injected directly into the tissue of the mammary gland of lactating goats.	123
28.	Effect of intravenous administration of emulsions of vitamin A palmitate, vitamin A alcohol and carotene to rats partially deficient in vitamin A.	134
29.	Levels of carotene and vitamin A in the blood plasma and milk fat after intravenous administration to a goat of a carotene emulsion.	135
30.	Recovery of carotene, xanthophylls and chlorophyll after blending with acetone and chromatographing on alumina.	174
31.	Recovery of tocopherol after adding to dried grass, blending with acetone and chromatographing on magnesia and alumina.	177
32.	The effect of various factors on the appearance of carotene and vitamin A in the blood plasma and livers of rats partially deficient in vitamin A 48 h after dosing orally with carotene.	209

## Table

## Page.

33. The effect of feeding separately ryegrass, low cyanide and high cyanide white clover and of feeding cyanide on the appearance of carotene and vitamin A in the blood plasma and liver of rats partially deficient in vitamin A. 210
34. The effect of ~~sub~~stantaneously administered ~~adrenaline~~ and insulin on the plasma vitamin A alcohol and ester levels of ~~stock~~ ~~colony~~ rats with normal and high vitamin A liver reserves. 211

## LIST OF FIGURES

Figure		Page
1.	Growth of completely deficient rats after injection of Tween dispersions of various carotenoids.	91
2.	Relationship between blood levels of tocopherol at the time of carotene injection and the quantity of vitamin A appearing in the liver of the rat 24 h later.	108
3.	Changes in the levels of various constituents in the milk fat of early- and late-calving, pasture-fed cows from early spring to late summer.	183
4.	Changes in the levels of various constituents in the blood plasma of early- and late-calving, pasture-fed cows from early spring to late summer.	184
5.	Changes in the levels of various pasture constituents throughout the season.	185
6.	Effect of reduced carotene intake and of supplementing with carotene in arachis oil on the level of carotene, xanthophyll, vitamin A alcohol and ester in milk fat.	198

LIST OF PLATES

Plate		Facing Page.
1.	Illustrating the technique of lung removal.	42
2.	Illustrating perfusion apparatus in operation.	53