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## RELATIONSHIP BETWEEN FAT COLOUR AND CAROTENOID AND RETINOL CONCENTRATIONS IN VARIOUS TISSUES AND SERUM IN CATTLE

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### INTRODUCTION

The adipose tissue of grazing cattle is usually a creamy-yellow colour. The yellow pigmentation results from the absorption of yellow carotenoids present in plant matter. A number of markets demand carcass-fat colour be white and, thus, those carcasses that do not reach specification are down-graded with considerable financial loss.

$\beta$ -Carotene is the major carotenoid absorbed in the small intestine (Palmer and Eckles, 1914) where some is converted to retinol (Moore, 1957). The remainder is transported and deposited intact in various tissues, particularly liver and adipose tissue (Moore, 1957). Carotene 15, 15'-dioxygenase is responsible for the splitting of  $\beta$ -carotene to retinol in intestinal mucosal cells and in liver (Goodman and Huang, 1965). The importance of this enzyme in determining fat colour has not been established, but is of considerable interest as retinol is a colourless product. It is known that sheep, compared with cattle, do convert more  $\beta$ -carotene to retinol as indicated by their high reserves of retinol in the liver and by the high dioxygenase activity in preparations from the intestinal mucosa (Yang *et al.*, 1992). Therefore it is argued that those animals able to convert more  $\beta$ -carotene to retinol may have less  $\beta$ -carotene deposited in their fatty tissues. The experiment described here was performed to investigate the variation in fat colour between individuals raised and fed on the same pasture and to establish whether or not a relationship exists between fat colour and  $\beta$ -carotene splitting.

### MATERIALS AND METHODS

A uniform group of 287 head of Shorthorn steers (approximately 500kg live weight) that had been grazed together on the Coopers Creek flood plains, Queensland for at least 12 months, were transported to an abattoir for slaughter. Samples of blood, liver and subcutaneous fat (rump) were collected serially from 100 carcasses on the slaughter line. All samples were returned to the laboratory within two hours. Subcutaneous fat samples were maintained at about 23 to 25°C until an objective assessment of fat colour had been performed using Minolta Chromameter, Model CR-200. Samples were then ranked in order of their  $b^*$  values and the fat samples having the lowest ( $n=26$ ) and those having the highest ( $n=24$ ),  $b^*$  values were selected as groups with 'white' or 'yellow' fat colour. Serum was prepared by centrifuging (1500g for 25 minutes) the blood which had been collected in plain glass tubes. Serum and tissue samples were stored frozen (-20°C) until required.

Carotenoid and retinol concentrations were measured on all selected samples of serum, fat and liver using high performance liquid chromatography (Yang *et al.*, 1992).

The mean carcass weight for all steers was 315kg with a mean fat thickness (P8) of 13 to 17mm. Eighty-five per cent of all steers had a dentition score of 6 to 8 teeth. The abattoir downgraded 8% of the carcasses for excessive yellow fat.

### RESULTS AND DISCUSSION

Objective assessment of subcutaneous fat from all cattle sampled ( $n=100$ ) indicated a wide range of fat colour within

the group of cattle, despite the fact that they were of similar breed, age and had been grazing the same pasture. Minolta  $b^*$  values of fat varied from 9.4 to 25.5. There was a high correlation between the total carotenoids of adipose tissue and Minolta  $b^*$  values ( $r=0.85$ ,  $n=50$ ,  $P<0.001$ ). The concentration of total carotenoids in individual samples of adipose tissue ranged from 0.39 to 3.47  $\mu\text{g/g}$  tissue.

Relationship between the concentrations of liver retinol and adipose tissue  $\beta$ -carotene. Although the activity of the  $\beta$ -carotene-splitting enzyme is regulated so that excessive amounts of retinol are not formed, the present work allows consideration of the possibility that when this activity is high, there would be less  $\beta$ -carotene for transport to, and deposition in, various tissues. Should this be the case, it would be expected that there would be a strong negative correlation between liver retinol and adipose tissue  $\beta$ -carotene concentrations ( $r=0.35$ ,  $n=50$ ,  $P<0.05$ ) was observed, suggesting that the activity of the dioxygenase in the intestinal mucosa does not play a significant role in the development of adipose tissue colour.

Other correlations. Significant correlations for all the tissue concentrations measured are given in Table 1. Apart from a highly significant relationship between the concentration of total carotenoids in the liver and serum ( $r=0.51$ ,  $n=50$ ,  $P<0.001$ ) there was essentially no other strong correlation. However, the concentration of retinol in the liver was related to the amount present in adipose tissue ( $r=0.39$ ,  $n=50$ ,  $P<0.05$ ). There was also a relationship between carotenoid concentrations in liver and adipose tissue ( $r=0.34$ ,  $n=50$ ,  $P<0.05$ ). A weak relationship was also apparent between liver retinol and adipose tissue carotenoids ( $r=0.35$ ,  $n=50$ ,  $P<0.05$ ).

'White' and 'yellow' fat colour groups. Treating the experimental samples as two individual groups ('white' and 'yellow' fat colour) proved to be useful for ascertaining various tissue concentrations for a large group of animals where dietary intake was essentially similar. Table 2 shows the mean values ( $\pm$  standard errors) for the 'white' and 'yellow' fat colour groups.  $\beta$ -Carotene was the predominant carotenoid in fat, accounting for 77.5% and 81.4% of the total carotenoids in fat of the 'white' or 'yellow' fat-colour groups respectively. Although the adipose tissue from the two groups differed markedly in their  $b^*$  values,  $\beta$ -carotene, lutein and total carotenoid concentrations ( $P<0.001$ ), the concentration of retinol in the tissues was the same for both groups, even though its concentration was about two to three times higher than that in serum. The concentration of both total carotenoids and retinol in liver was significantly greater in the 'yellow' fat-colour group ( $P<0.05$ ).

Retinol concentrations in serum from the two groups was not significantly different. Serum concentrations of total carotenoids were also similar in the 'white' and 'yellow' fat-colour groups, confirming that the cattle had been grazed on the same pasture. This finding is of considerable interest as it demonstrates that those cattle in the 'white' fat-colour group do not accumulate as much carotenoid even though their fat tissues are perfused with similar concentrations in the serum. This may suggest that a specific mechanism exists in adipose tissue which controls the uptake of carotenoids. We are presently investigating the uptake of  $\beta$ -carotene by isolated adipocytes and the binding of  $\beta$ -carotene to adipocyte plasma membranes.

This study was undertaken assuming that these animals had the same carotenoid intake and absorbed the same amount of carotenoids. It was also assumed that the conversion to retinol by other tissues, such as the liver (Moore, 1957) was minimal and therefore, did not affect the retinol content in the liver. Despite all these limitations, it can be concluded from this study that, as far as the colour of body fat is concerned, the carotene-splitting activity in the intestinal mucosa plays only a secondary role in the metabolism of carotenoids. It is likely that it is individual differences in the selective absorption process in the small intestine, and/or the uptake of  $\beta$ -carotene by the adipose tissue, that are responsible for the different fat colour observed in animals.

#### ACKNOWLEDGMENT

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Table 1. Significant ( $P < 0.05$ ) correlations between serum, subcutaneous fat and liver in their carotenoid and retinol concentrations.

	Fat				Liver total C
	b* value	$\beta$ -carotene	Total C <sup>1</sup>	Retinol	
Total (n=50)					
Fat $\beta$ -carotene	0.84				
Total C	0.85	0.99			
Liver Total C		0.36	0.34		
Retinol	0.29	0.34	0.35	0.39	
Serum Total C					0.51
'White' fat colour (n=26)					
Fat $\beta$ -carotene	0.52				
Total C	0.49	0.99			
Liver Total C		0.39			
Retinol		0.48	0.51		
Serum Total C					0.57
'Yellow' fat colour (n=24)					
Fat $\beta$ -carotene	0.44	0.97			
Total C	-0.51				
Liver Total C					
Retinol				0.47	
Serum Total C					0.44

<sup>1</sup> Total carotenoids.

Table 2. Carotenoid and retinol concentrations in serum, subcutaneous fat and liver (Mean±SE).

	Fat colour		Signif. <sup>1</sup>
	'White' n=26	'Yellow' n=24	
Subcutaneous fat (µm/g)			
β-carotene	0.86 ± 0.060	1.79 ± 0.083	***
Total C <sup>2</sup>	1.11 ± 0.072	2.20 ± 0.093	***
Retinol	0.97 ± 0.063	1.00 ± 0.093	NS
b* value	13.2 ± 0.29	21.2 ± 0.29	***
Liver (µm/g)			
Total C	9.05 ± 0.510	10.56 ± 0.418	*
Retinol	192 ± 13.6	255 ± 23.2	*
Serum (µm/g)			
Total C	2.10 ± 0.149	2.27 ± 0.162	NS
Retinol	0.44 ± 0.017	0.42 ± 0.013	NS

<sup>1</sup> Statistical significance: NS=not significant (P>0.05); \* P<0.05; \*\*\* P<0.001.

<sup>2</sup> Total carotenoids.