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Vitamin A and colour parameters in pig fat as possible biomarkers of feeding traceability

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1. INTRODUCTION

Consumers

Safe & High Quality Food Products

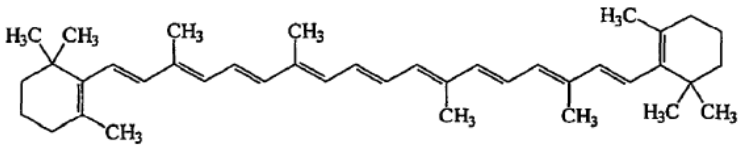
TRACEABILITY

Animal Products Traceability

Different constituents in feedstuffs appear in the animals' blood and tissues upon consumption

Carotenoids as biomarkers of grazing animals in other species

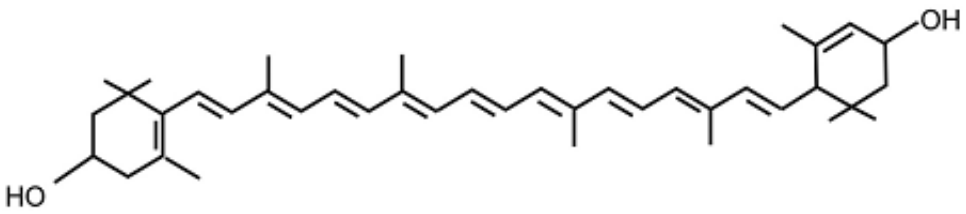
(few information available about pigs)

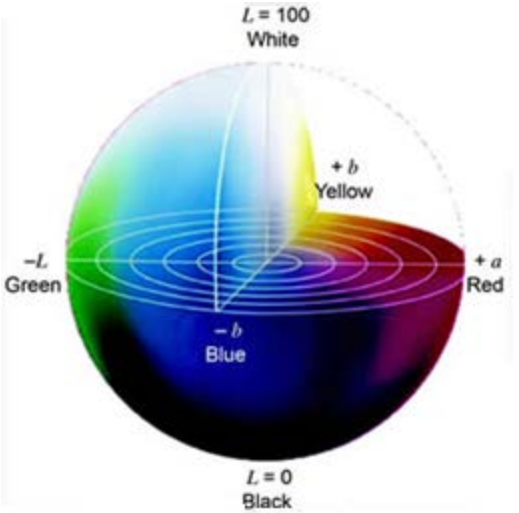


Colour parameters and traceability

Carotenoids in adipose tissue → Differences in carcass fat colour

Are colour parameters of the CIELab space useful for traceacbility purposes?





2. AIM OF THE STUDY

Assess the usefulness of carotenoids, vitamin A (retinol) and colour parameters in perirenal fat to differentiate pigs with different types of diet, in order to evaluate their validity as biomarkers of traceability.

3. MATERIALS & METHODS

- Animals and diets

45 animals divided in 3 groups (3x15)

Group 1:
- 14 month-old 100% Iberian breed pigs (150 ± 3 kg).
- Montanera feeding system: acorns + pasture

Group 2:
- 14 month-old 100% Iberian breed pigs (161 ± 2 kg).
- Indoors: concentrate.

Group 3:
- 6 month-old commercial crossbred pigs (88 ± 1 kg).
- Indoors: concentrate.

Montanera feeding system

Dehesa forest: Acorns and grass (4 months)

Slaughter later → deposit enough intramuscular fat → good fat infiltration

Exercise → meat quality

Sampling

Representative samples of the different diets from the farms (Southwest of Spain).

→ Carotenoids

Perirenal fat at the moment of the slaughter

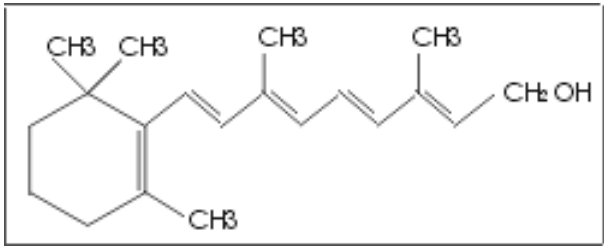
→
- Carotenoids & Retinol
- Colour measurements

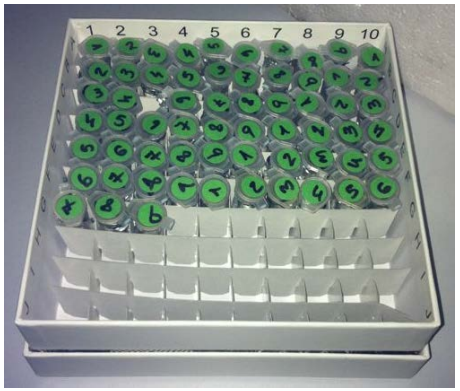
- Samples were stored at -80ºC.
- Previously to the analysis, all the samples were unfrozen overnight and in the dark in a fridge at 4ºC.

- Carotenoids and Retinol extractions

o Food samples

5 g + hexane/ethanol → saponification (15% KOH solution) → washing → drying → + ethyl acetate → HPLC





o Fat samples

500 mg + 1 ml BHT→ saponification (30% KOH solution) → ether/hexane → washing → drying → + ethyl acetate → HPLC

- HPLC conditions
  - Agilent 1100 system.
  - YMC C_{30} (feedstuffs) & C_{18} (fat extracts).
  - Mobile phase: methanol + methyl *tert*-butyl ether + water
  - 325 nm (Retinol)
  - 450 nm (Carotenoids)

- Instrumental colour parameters

  - CIELab space (CIE, 1986).
  - C^*_{ab}, L*, a*, b* and h_{ab}
  - Spectrocolorimeter CM-700d (Konica Minolta Holdings, Inc, Osaka, Japan)
  - D_{65} Illuminant
  - 10º Observer
  - Zero and white calibration
  - One hour after the slaughter.

4. RESULTS

Group	Feeding	Violaxanthin	Zeaxanthin	Lutein	ß-carotene
1	Pasture	71.70 ± 7.07	n.d.	169.79^{a} ± 14.59	242.93 ± 22.45
2	Concentrate	n.d.	5.58 ± 0.22	9.53^{b} ± 0.41	n.d.
3	Concentrate	n.d.	3.93 ± 0.85	6.39^{b} ± 1.57	n.d.
	Significance	-	n.s.	**	-

Means (mg/100 g), standard error, ANOVA and multiple comparison Tukey test for the carotenoids identified in the feedstuffs: group 1 (Iberian pigs fed on montanera system), group 2 (Iberian pigs fed on concentrate), group 3 (commercial pigs fed on concentrate)
n.d.: not detected

Variable	Group 1	Group 2	Group 3	Significance
mg Retinol/g fat	$4.09^{a} \pm 0.36$	$3.69^{a} \pm 0.22$	$6.02^{b} \pm 0.28$	***
L*	$57.56^{a} \pm 1.89$	$69.55^{b} \pm 2.66$	$74.30^{b} \pm 0.81$	***
a*	$2.36^{a} \pm 0.46$	$0.94^{a} \pm 0.54$	$4.51^{b} \pm 0.55$	***
b*	$9.97^{a} \pm 0.64$	$8.36^{a} \pm 0.54$	$14.34^{b} \pm 0.62$	***
C_{ab}	$10.49^{a} \pm 0.68$	$8.69^{a} \pm 0.61$	$15.14^{b} \pm 0.68$	***
h_{ab}	$81.52^{ab} \pm 2.77$	$87.13^{b} \pm 2.91$	$72.81^{a} \pm 1.74$	**

Mean values, standard error, ANOVA and multiple comparison Tukey test for the retinol levels in fat and colour parameters measured in the fat of the three groups of animals: group 1 (Iberian pigs fed on montanera system), group 2 (Iberian pigs fed on concentrate), group 3 (commercial pigs fed on concentrate)
n.d.: not detected

Retinol

Group	Predicted group (%) 1	2	3
1	**20.8**	41.7	37.5
2	29.2	**58.3**	12.5
3	13.8	10.3	**75.9**

Retinol concentration in adipose tissue → **53.2 %**

Genotype & Age

Colour parameters

Group	Predicted group (%) 1	2	3
1	**84.4**	15.6	0
2	37.5	**62.5**	0
3	0	6.7	**93.3**

L^*, a^*, b^*, C_{ab}, h_{ab} measured in adipose tissue → 78.9 %

Diet

5. CONCLUSIONS

- Retinol contents in renal fat was significantly different ($p<0.001$) in commercial (group 3) and Iberian breed pigs (groups 2 and 3), being more related to age or genotype than to the type of diet.

- L* seemed to be a good parameter to differentiate the animals according to the diet: Iberian pigs reared on montanera feeding system were significantly different ($p<0.001$) from the other two groups.

- From a traceability point of view, colour parameters in renal fat could be proposed to differentiate pigs according to their diet.

THANK YOU FOR YOUR ATTENTION