Meat Science 122 (2016) 132–138

Contents lists available at ScienceDirect

Meat Science

journal homepage: www.elsevier.com/locate/meatsci

Calculating the iodine value for Italian heavy pig subcutaneous adipose tissue from fatty acid methyl ester profiles

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ARTICLE INFO

Article history:
Received 7 April 2016
Received in revised form 13 July 2016
Accepted 3 August 2016
Available online 06 August 2016

Keywords:
Heavy pig
Fatty acids
Iodine value
Estimating
Regression equations

ABSTRACT

In this work, different equations were compared as for their effectiveness in predicting the iodine value (IV), based on fatty acid (FA) composition of subcutaneous adipose tissue of Italian heavy pigs. In particular, six equations were tested: AOCS (1); modified AOCS (2), including all unsaturated FA (UFA); regression models obtained using the stepwise regression procedure as variable selection method, calculated considering only UFA (3) or all the FA (4); regression models obtained using the backward elimination procedure, calculated considering only UFA (5) or all the FA (6). The comparison of the equations performance, estimated using an external test set, showed that the use of regression models led to significant enhancements of prediction accuracy with respect to the AOCS equations. Using both equations 4 and 6, the average paired differences between experimental and predicted IV values were not statistically significant. Therefore, it is possible to use these equations for IV estimation of the subcutaneous adipose tissue of Italian heavy pigs.

1. Introduction

In pig production, the fat components of the carcass play a role of considerable importance, both from a nutritional point of view, and consequently for the acceptability by the consumer, and under the technological aspect (Whittington, Prescott, Wood, & Enser, 1986). For Protected Designation of Origin (PDO) products, the Italian processing industry requires carcasses from pigs slaughtered at heavy weights (160–170 kg), belonging to genetic types under the control of the national selection or from commercial hybrids recognized as suitable for these types of production (Lo Fiego, 1996; Lo Fiego, Santoro, Macchioni, & De Leonibus, 2005).

Even in Italy, as in other European countries (Andersen, 2000), to meet the needs of the consumer, the selection has been oriented toward the reduction of fat depots of carcasses, resulting in higher meatiness (Lo Fiego, 1996). This reduction caused an increase in the level of unsaturated lipids mainly linoleic acid (Gandemer, 2002; Lo Fiego, Macchioni, Minelli, & Santoro, 2010; Pettigrew & Esnaola, 2001; Piedrafita, Christian, & Lonergan, 2001). A high content of polyunsaturated fatty acids may cause troubles problems in meat processing, namely concerning the consistency of the meat products and their resistance to oxidative phenomena (Houben & Krol, 1983; Santoro, 1983). For the Italian PDO production a white fat is required, with optimal consistency and high oxidative stability: these parameters are the main quality characteristics of fat (Wood, 1984) and are closely monitored by the Consortia of PDO hams. In order to reduce the occurrence of problems during the curing period and to ensure an excellent quality of the finished products, some threshold values were fixed in the production rules of the main Italian PDO hams, for fat thickness, iodine value (IV) and linoleic fatty acid content (C18:2) (Modena: MIPAF, 1999; Parma: Consorzio del Prosciutto di Parma, 1992; San Daniele: MIPAF, 2007). The fat thickness of the subcutaneous adipose tissue of the thigh cannot be <15 mm (optimum 20–30 mm) and the IV and the C18:2 content of the lipids must not be higher than 70 and 15%, respectively.

The methods for evaluating the quality of fat are manifold, ranging from subjective methods (Wood, 1984), usually considered unreliable (Enser, 1983), to objective methods, primarily aimed to determine or estimate the fatty acid composition and texture of fat tissue (Foca et al., 2013; Foca et al., 2016; García-Olmo et al., 2002; Olsen, Baustad, Egelandsdal, Rukke, & Isaksson, 2010; Pérez-Marín, De Pedro Sanz, Guerrero-Ginel, & Garrido-Varo, 2009; Seman, Barron, & Matzinger, 2013; Zudaire & Alfonso, 2013).

The methods used by the control systems of Italian PDO are mostly based on the direct evaluation of the fatty acid composition of lipids by gas chromatography and on indirect assessment of the overall degree of unsaturation by determination of IV. The latter is

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http://dx.doi.org/10.1016/j.meatsci.2016.08.002
0309-1740/

Table 1
Carcass characteristics and lipid fatty acid composition of the subcutaneous adipose tissue (average values ± standard deviation).

Parameter	Units	Training set (n = 261)	Test set (n = 103)
Carcass weight	kg	131.05 ± 10.1	132.70 ± 10.4
Backfat thickness[a]	mm	30.44 ± 7.72	27.14 ± 5.80
Raw thigh weight	kg	16.54 ± 1.18	16.50 ± 1.44
Fatty acids composition	%		
Myristic (C14:0)	"	1.34 ± 0.15	1.38 ± 0.25
Palmitic (C16:0)	"	23.76 ± 1.15	24.07 ± 1.80
Margaric (C17:0)	"	0.32 ± 0.10	0.42 ± 0.17
Stearic (C18:0)	"	13.12 ± 1.47	14.04 ± 1.34
Arachidic (C20:0)	"	0.20 ± 0.07	0.20 ± 0.05
Total SFA	"	**38.74 ± 2.20**	**40.11 ± 2.94**
Palmitoleic (C16:1)	"	2.15 ± 0.39	2.01 ± 0.41
Eptadecenoic (C17:1)	"	0.28 ± 0.12	0.38 ± 0.17
Oleic (C18:1)	"	43.15 ± 2.20	41.94 ± 2.47
Eicosenoic (C20:1)	"	0.90 ± 0.16	0.95 ± 0.18
Total MUFA	"	**46.48 ± 2.40**	**45.28 ± 2.75**
Linoleic (C18:2)	"	13.14 ± 2.74	13.06 ± 4.25
α-Linolenic (C18:3)	"	0.69 ± 0.21	0.66 ± 0.26
Eicosadienoic (C20:2)	"	0.62 ± 0.13	0.60 ± 0.19
Eicosatrienoic (C20:3)	"	0.15 ± 0.06	0.12 ± 0.06
Arachidonic (C20:4)	"	0.18 ± 0.06	0.17 ± 0.07
Total PUFA	"	**14.78 ± 3.08**	**14.61 ± 4.71**
Unsaturation coefficient[b]		1.26 ± 0.05	1.26 ± 0.07

SFA: saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids.
[a] Average of two measurements carried out respectively between the 3/4 last lumbar vertebra and 3/4 last rib at 8 cm from the splitting line of the carcass.
[b] Unsaturation coefficient = ∑(% of each unsaturated fatty acid × number of its double bonds) / % unsaturated fatty acids.

a chemical parameter currently used for the evaluation of chemical-physical characteristics of the fat (Berhe et al., 2016; Seman et al., 2013) and numerous authors, as reported by Hugo and Roodt (2007), have established the maximum limits in the range from 60 to 75, to ensure optimum characteristics of the lipids in the processed products.

Among the different methods for measuring IV, the Wijs method (AOAC, 1984) is the most widely used one (Kyriakidis & Katsiloulis, 2000). This method, however, is time consuming, it requires a significant amount of sample and the use of hazardous chemicals (Kyriakidis & Katsiloulis, 2000; Seman et al., 2013). Therefore, for control systems and for research purposes, the theoretical IV is often calculated, based on the fatty acid composition of lipids determined by gas chromatography (American Oil Chemists' Society (AOCS), 1998, 2009; International Organization for Standardization (ISO), 2013).

Nowadays, the modern gas chromatographic equipment allows to determine the fatty acid composition of lipids in short times (Ficarra, Lo Fiego, Minelli, & Antonelli, 2010; Ichihara, Shibahara, Yamamoto, & Nakayama, 1996), which in turn makes it possible to obtain a fast estimate of IV.

Many authors (Asmus et al., 2014; Benz et al., 2011; Hallenstvedt, Øverland, Rehnberg, Kjos, & Thomassen, 2012; Musella et al., 2009; Nemechek et al., 2015; Wiegand, Hinson, Ritter, Carr, & Allee, 2011) calculated IV of the fat depots of pig carcasses according to the following AOCS Cd 1c-85 (1998) equation:

$$IV = 100 \times \sum \frac{Af \times 253.81 \times db}{MWf}$$

where IV is the iodine value, *Af* is the amount (%) of each fatty acid in the mixture, *db* is the number of its double bounds, 253.81 is the atomic weight of two iodine atoms and *MWf* is the molecular weight of the fatty acid methyl ester (FAME) in the triglyceride form (Knothe, 2002; Pétursson, 2002).

In detail, equation AOCS Cd 1c-85 (1998) includes the following terms:

$$IV = [C16:1] \times 0.950 + [C18:1] \times 0.860 + [C18:2] \times 1.732 + [C18:3] \times 2.616 + [C20:1] \times 0.785 + [C22:1] \times 0.723 \quad (AOCS) \qquad (1)$$

where the fatty acid composition (%) is in brackets.

This formula includes only some of the fatty acids present in the lipids (Pétursson, 2002) and it is intended for general use, i.e., it could be applied to any type of plant oils (Kyriakidis & Katsiloulis, 2000). However, the authors pointed out that the type of considered oil can influence its accuracy, and the results of studies on the correlation between AOCS and Wijs methods were not always satisfactory. For fish oil, Ham, Shelton, Butler, and Thionville (1998), while reporting statistically significant differences between the two methods, considered however satisfactory the concordance, therefore suggesting that the AOCS method could be used as the official method for calculating IV from the fatty acid composition. Kyriakidis and Katsiloulis (2000), in a comparison between the two methods, found statistically significant differences for olive, corn, soybean and sunflower oils, and consequently proposed different equations for these oils.

The literature does not report data for comparison between the Wijs method and the AOCS method relatively to pig fat. The purposes of this study were: i) to compare the IV determined by the Wijs method with that calculated by the formula proposed by AOCS (1998) (AOCS) and ii) to check any variations determined by the integration of this formula with the inclusion of all the unsaturated fatty acids detected by gas chromatography (AOCS_1, see below). Furthermore, the study aimed to verify the possibility of developing specific equations for the calculation of the IV of the lipids of the subcutaneous adipose tissue of Italian heavy pigs, starting from the fatty acid composition of lipids determined using capillary gas chromatography.

2. Material and methods

2.1. Animals and sampling procedure

The sample examined and used for the development of new estimating equations (training set) consisted of 261 left thighs coming from carcasses of heavy pigs (average weight of carcass 131.0 ± 10.1 kg), randomly selected from genetic types commonly used and representative of the production of Italian heavy pigs. The calculated equations have been then tested on an independent sample (external test set) of 103 left thighs of carcasses equally obtained from Italian heavy pigs (average weight of carcass 132.7 ± 10.4 kg), coming from different slaughtering batches and farms with respect to those considered in the training set. This allowed to perform a severe evaluation of the actual predictive capabilities of the obtained regression models by simulating their performances in completely independent conditions. After slaughter, during grading of the carcasses, individual carcass weight was recorded and backfat thickness between the 3/4 last lumbar vertebra and 3/4 last rib, at 8 cm from the splitting line of the carcass, was measured by Fat-o-Meater. After carcass cutting, the left thigh of each carcass was weighed, chilled at 0–4 °C for 24 h and then, at trimming, a sample of

Table 2
Observed (Wijs, A) vs. calculated (AOCS, B, and AOCS_1, C) iodine value (IV).

Data set	(A): Wijs	(B): AOCS	(C): AOCS_1	Differences[a]	
				A − B	A − C
Training[b]	67.27 ± 4.60	64.36 ± 4.06	66.52 ± 4.34	2.91[c]	0.75[c]
Test[b]	66.46 ± 4.91	63.02 ± 6.35	65.14 ± 6.65	3.44[c]	1.32[c]

[a] Paired *t*-test.
[b] Mean ± standard deviation.
[c] P < 0.0001.