PROC 25th EUROP. MEETING OF MEAT RES. WORKERS BUDAPEST, vol 2 pp 767-772

A alternative method for detection of beef tallow in pork fat

ROGER VERBEKE and HUBERT F. DE BRABANDER
Laboratory of Chemical Analysis of Food of Animal Origin, Veterinary Faculty of the University of Ghent, Belgium

The incorporation of palmitic acid or oleic acid + linoleic acid in the 2-position of the triglycerides of pork and beef fat are shown to be highly correlated to its corresponding acid contents of the triglycerides. On basis of the different regressions obtained for pork and beef fat, the percentage adulteration of pork fat with beef tallow may be determined.

The method involves gaschromatographic determination of the triglyceride fatty acids and analysis of the fatty acid content in the monoglycerides obtained after lipase treatment of the fat. The results indicate that mixing pig fat with 5 % beef fat, or mixing beef fat with 10 % pig fat may be accurately estimated. This method is superior to the existing methodologies in detecting adulteration of pork fat with beef tallow.

Eine Alternativmethode zum Nachweis von Rinderfett in Schweineschmalz

ROGER VERBEKE und HUBERT F. DE BRABANDER
Laboratorium für chemische Analysen von Nahrungsmitteln tierischen Ursprungs, veterinärmedizinische Fakultät der Universität Gent, Belgien

Es wird gezeigt, daß die Aufnahme von Palmitinsäure oder Ölsäure + Linolsäure in 2-Position der Triglyceride von Schweine- und Rinderfett vollkommen mit dem entsprechenden Säuregehalt der Triglyceride korreliert. Auf der Basis der verschiedenen, bei Schweine- und Rinderfett erhaltenen Regressionswerte wurde der Prozentsatz der Verfälschung von Schweineschmalz durch Rindertalg bestimmt.

Die Methode umfaßt eine Analyse der Fettsäuren der Triglyceride durch Gaschromatographie und eine Bestimmung des Fettsäuregehalts der Monoglyceride, die nach Behandlung des Fettes mit Lipase erhalten wurden. Die Ergebnisse zeigen, daß eine Zumischung von 5 % Rindertalg zu Schweineschmalz, oder umgekehrt 10 % Schweineschmalz zu Rindertalg genau nachgewiesen werden kann. Diese Methode ist den vorhandenen Methoden zum Nachweis der Verfälschung von Schweineschmalz durch Rindertalg überlegen.

9.3

Méthode pour la détection de graisse de boeuf dans le saindoux

ROGER VERBEKE et HUBERT, F. DE BRABANDER
Laboratoire d'analyses chimiques des denrées alimentaires d'origine animale, Faculté de Médecine Vétérinaire, Université de l'Etat à Gand, Belgique

L'incorporation de l'acide palmitique ou des acides oléïque + linoléïque en position 2 des graisses de porc et de boeuf a été montrée en parfaite corrélation avec leur teneur respective dans les triglycérides. Sur la base des différentes régressions obtenues à partir des graisses de porc et de boeuf, le pourcentage d'adultération du saindoux par le suif fut déterminé.
La technique comprend une analyse des acides gras des triglycérides par chromatographie gazeuse et une détermination des acides gras des monoglycérides obtenus après traitement des graisses avec du lipase. Les résultats témoignent qu'une admixtion de 5 % de suif au saindoux, ou inversement de 10 % de saindoux au suif est déterminable avec précision.
La méthode décrite est supérieure aux méthodologies existantes dans la détection de l'adultération du saindoux par le suif.

Метод обнаружения говяжьего жира в свином жире

РОЖЕ ВЕРБЕКЕ и ГУБЕРТ Ф. ДЕ БРАБАНДЕР
Лаборатория химического анализа продуктов питания животного происхождения, Ветеринарный факультет, Гентский Государственный Университет, Бельгия.

Было показано, что введение пальмитиновой кислоты и кислот олеиновой + линолевой в 2-положение говяжьего и свиного жиров проявляет тесную корреляцию с содержанием триглицеридов. На основе различных регрессий, полученных по свиному и говяжьему жирам, получается процент говяжьего жира, фальсифицирующего свиной жир.
Применение метода включает анализ жирных кислот путём газовой хроматографии в триглицеридах и определение жирных кислот в моноглицеридах, полученных по обработке жира липазой. Результаты свидетельствуют о том, что добавка 5% говяжьего жира к свиному, или, наоборот, 10% свиного к говяжьему может определяться точно. Описанный метод превосходит существующие методы обнаружения примеси говяжьего жира в свином.

An alternative method for the detection of pork fat adulteration with beef tallow

ROGER VERBEKE and HUBERT, F. DE BRABANDER

Laboratory of Chemical Analysis of Food of Animal Origin, Veterinary Faculty of the University of Ghent, Belgium

Introduction

Adulteration of pig lard with beef or lamb fat is normally detected by the decrease of the Boemer value. However, determination of the Boemer value is time consuming and requires skilled analysts to obtain sufficiently repeatable results. However the method is rather insensitive to mixing of lard with transesterified fats or low melting beef tallow. Recently, we showed that the different fatty acids incorporated in position 2 of beef and pork fat were closely correlated to the corresponding fatty acid contents in the total triglycerides (7). This aspect of the work has been extended to support the conclusion that this relationship can be used to determine quantitatively the percent adulteration of pork fat with beef tallow.

Material and Methods

At least 10 samples of backfat or kidney fat were sampled in the slaughterhouse from oxen, steers or heifers originating from different farms. Ten samples of backfat were obtained on different farms from sows or barrows. Each sample was homogenised, molten and filtered at 80 °C. The clear fat was stored in the freezer (- 20 °C) until used. Fats were transesterified by incubating 20 mg fat in presence of 1 ml sodium methylate solution (0.025 N) in methanol at 90 °C during 1 hr. The fatty acid composition in position 2 of the triglycerides was determined by a modification of the method of DUTTA et al. (3). Pancreaslipase (30 mg ; E.C. n° 3.1.1.3 ; Sigma type II) was homogenised with 1 ml 1M TRIS-buffer (pH = 8) containing 25 µl 40 % $CaCl_2$ solution. On silicagelplates (10 x 20 cm) a 2 x 8 cm lipase reactionband was formed by application of 200 µl lipase solution. The plate was dried under a cold air stream in order to remove most of the water and 100 µl of a fat solution (80 mg of fat in 1 ml n. hexane) was evenly applied over the lipase reactionband. The silicagel plate was placed immediately in a waterbath (40 °C) with the silicagellayer situated at +2 cm above the water surface. After 10 min. incubation the plate was removed and dried. The lipid mixture was concentrated into a narrow band by developping the plate three times with diethyl ether over a distance of ±5 cm. The lipase reaction band was removed by cutting off that part of the plate. The remainder of the plate was developped in n. hexane-diethyl ether (1 : 1, v/v) over a distance of 12 cm. After drying, the different fractions were visualised with iodine vapour. The monoglyceride fraction was transferred into a small column (0.6 mm I.D.) and elution was performed with 2, 1 and 1 ml freshly distilled, dry diethyl ether. The ether was evaporated under a jet of nitrogen. The lipids were transesterified with 200 µl sodiummethylate solution.

The gaschromatograph used was a Hewlett-Packard 5750. A capillary column (100 m ; 0.5 mm I.D. R.S.L., Belgium) coated with Silar 10 C was used. The carriergas was H_2 at 5 ml/min. The temperature of the column, the injector and the detector was at 180, 210 and 220 °C respectively.

Results and discussion

1. Distribution of fatty acids within the triglycerides of lard and beef tallow

The fatty acid distribution observed in 10 samples of lard and 10 samples of beef tallow are given in Table 1. In agreement with earlier reports (4,5), palmitic acid (C16:0) of pork fat is mainly incorporated in the 2-position while oleic acid and linoleic acid (C18:(1+2)) are preferentially esterified in the 1 and 3 positions of the triglycerides of lard. As in most mammalian fats, the unsaturated fatty acids in beef fat tend to prefer position 2 while palmitic acid accumulates in positions 1 and 3. The large variability in the proportions of the fatty acids incorporated in position 2 of the triglycerides do not allow to detect an adulteration of less than 30 % of beef tallow in lard.

Recently, we showed that the incorporation of different fatty acids in position 2 from beef or pork fat was closely correlated to their corresponding contents in the total triglycerides (7). From our results and including literature data, close correlations between the amount of C18:(1+2) incorporated into position 2 and the total C18:(1+2) content in the triglycerides of lard and beef was observed (Fig. 1). The slopes of the regression lines of beef and lard were parallel but showed characteristic differences in elevation ($p < 0.001$). Analogous correlations ($p < 0.001$) between the amount of palmitic acid incorporated in position 2 and the total palmitic acid content of beef and lard have been observed for beef tallow and pork fat (7).

2. Quantitative determination of adulteration of lard with beef tallow

Since the regression lines, showing the incorporation of C18:(1+2) into pos. 2, run parallel for lard and beef tallow, fat adulteration is revealed by differences in elevation. Similarly a linear discriminating function for beef tallow and lard was calculated from the regressions

Table 1.

Mean fatty acid composition (mole %) of whole triglycerides and of fatty acids at the 2-position of lard and beef tallow.

Fatty acid	Mean fatty acid content ± standard error					
	C14:0	C16:0	C16:1	C18:0	C18:1	C18:2
Lard (n = 10)						
in triglyceride	1.9 ± 0.2	29.4 ± 1.2	3.2 ± 0.4	14.1 ± 1.8	42.2 ± 2.6	9.2 ± 1.4
in 2-position	4.6 ± 0.7	70.6 ± 3.5	4.3 ± 0.9	5.2 ± 1.1	12.0 ± 2.2	2.6 ± 1.0
proportion in 2-position[a]	82 ± 17	80 ± 3.7	45 ± 11	12.4 ± 2.8	9.4 ± 1.5	10.3 ± 2.2
Beef tallow (n = 10)						
in triglyceride	4.2 ± 0.4	31.7 ± 2.7	1.6 ± 0.6	27.9 ± 4.3	31.2 ± 3.4	1.3 ± 0.5
in 2-position	8.3 ± 0.9	22.6 ± 2.3	3.0 ± 1.4	18.1 ± 2.8	44.4 ± 4.9	2.0 ± 0.9
proportion in 2-position[a]	66 ± 7	23.9 ± 2.8	62 ± 5	21.6 ± 2.0	47.6 ± 3.8	48.6 ± 19.0

[a] $\text{proportion in 2-position} = \frac{\text{mole \% in 2-position}}{3 \times \text{mole \% in triglyceride}} \times 100$

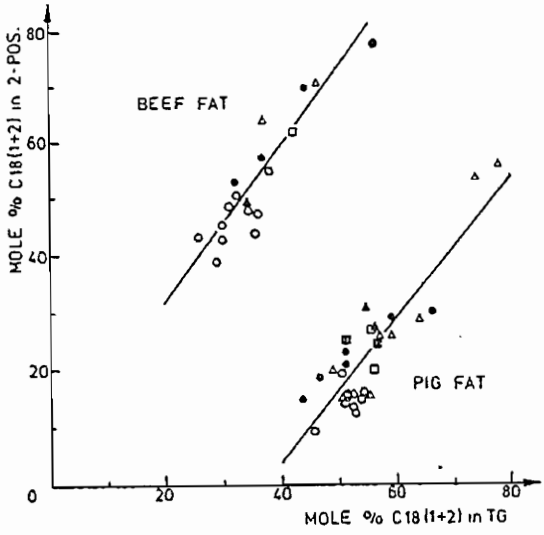

Fig. 1. Relationship between incorporation of oleic acid + linoleic acid in the 2-position (=Y) of beef and pork fat and unsaturated fatty acid content (=X).

Beef fat : $Y = 1.40\ X + 3.65$ $r = 0.90^a$

Pig fat : $Y = 1.25\ X - 46.21$ $r = 0.88^a$

Data : ○this work; ●(2); ●▲(4); ▲(5); ■(1); □(8).

$^a p < 0.001$

observed for the C18:(1+2) content in position 2 of the triglyceride :

$U = 0.0694\ X - 0.0479\ Y$

where :

Y = C18:(1+2) in position 2 of the triglyceride

X = C18:(1+2) content of the total triglyceride

In applying this equation to the mean values of C18:(1+2) observed for lard (Table 1) the value U(lard) = 2.857. For beef the value U(tallow) = 0.010 was obtained.

The percentage adulteration of an unknown lard sample (Ux) may be calculated as :

Adulteration % =

$$\frac{U(lard) - Ux}{U(lard) - U(tallow)} \times 100 = \frac{2.857 - Ux}{2.847} \times 100$$

Calculation of the "adulteration" of 20 individual fats gave a mean value of 0.0 ± 4.3 % for pig fat and 100 ± 5.1 % for beef tallow (Table 2). Application of this method to 2 fats from "unsaturated" pigs, showing a divergent fatty acid composition, yielded values similar to the ones observed on our

Table 2.

Percentage adulteration of pork fat calculated from GLC-analysis of oleic acid and linoleic acid in the 2-position of the triglycerides.

	C18:(1+2) content (Mole %) in triglyceride	C18:(1+2) content (Mole %) in 2-position	Estimation of % adulteration of pork fat
Pure lard (n = 10)	51.4 ± 2.5	14.8 ± 2.5	0 ± 4.3
Unsaturated pig H	65.9	35.0	-1.4
Unsaturated pig V	77.0	56	+6.8
Pure beef tallow (n = 10)	32.2 ± 3.8	46.4 ± 4.5	100 ± 5.1
Mixtures beef tallow + lard			
2 + 98	51.7	18.6	+5.7
4 + 96	49.8	16.9	+7.2
8 + 92	50.1	18.4	+9.2
12 + 88	49.4	20.3	+14.2
20 + 80	48.5	22.7	+20.4

lard samples. The method was checked in melting increasing concentrations of beef tallow in pig fats and determining experimentally the C18:(1+2) composition of the triglycerides and the corresponding 2-monoglycerides. From our results it is evident that mixing 10 % beef tallow in lard may be correctly estimated (Table 2). Similar results are obtained with the relations between the incorporation of palmitic acid (7).

Conclusions

The results show that adulteration of pig fat with 10 % beef fat may be correctly estimated. This relatively simple analytical technique was found to be at least as sensitive as the Boemer method. Measurement of the incorporation of the fatty acids into the 2-position of the triglycerides has not the limitation of misclassification observed in the Boemer method with lard adulterated with low melting beef tallow or transesterified triglycerides (6,7).

Aknowledgements

The authors are grateful to Prof. KROL and Ir. HOUBEN (Utrecht, the Netherlands) for supplying the unsaturated pig fat samples.

References

1. Brockerhoff, H., Hoyle, R.J., Hwang, P. and Litchfield, C. (1968) Lipids 3, 24.
2. Coleman, M.H. (1961) J. Am. Oil Chem. Soc. 38, 685.
3. Dutta, J., Das, A.Kr., Saha, S. (1978) J. Chromatogr. 154, 39.
4. Hilditch, T.P. and Williams, P.N. (1964) "The chemical constitution of natural fats", 4th Ed. Chapman and Hall, London.
5. Mattson, F.H., Volpenhein, R.A. and Lutton, E.S. (1964) J. Lipid Res. 5, 363.
6. Roos, J.B. (1962) Fette, Seifen, Anstrichmittel 64, 303.
7. Verbeke, R. and De Brabander, H. (1979) Vl. Diergeneesk. Tdschr. 48, 47.
8. Youngs, C.G. (1961) J. Am. Oil Chem. Soc. 38, 62.