SN

中华人民共和国出入境检验检疫行业标准

SN/T 0526—2015
代替 SN 0526—1996

出口食品中增效醚残留量的检测方法 液相色谱-质谱/质谱法

Determination of piperonyl butoxide residues in foodstuffs for export—LC-MS/MS method

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前　言

本标准代替 SN 0526—1996 出口粮谷中增效醚残留量检验方法。

本标准与 SN 0526—1996 相比主要修改如下：

——修改了标准名称。

——删除了“抽样”部分。

——修改了样品的前处理方法。

——修改了仪器测定方法。

——增加了方法适用范围。

——修改了标准结构。

——降低了方法测定低限。

本标准按照 GB/T 1.1—2009 给出的规则起草。

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本标准由国家认证认可监督管理委员会提出并归口。

本标准起草单位：中华人民共和国河南出入境检验检疫局。

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出口食品中增效醚残留量的检测方法 液相色谱-质谱/质谱法

1 范围

本标准规定了食品中增效醚残留量的制样方法与液相色谱-质谱/质谱测定方法。

本标准适用于猪肉、鸡肝、牛肾、牛脂肪、鸡蛋、牛奶、柑桔、大豆、花生、大蒜、马铃薯、菠菜、芹菜、小麦、大米、大麦、芝麻等食品中增效醚残留量的液相色谱-质谱/质谱的测定。

2 规范性引用文件

下列文件中的条款通过本文件的引用而成为本文件的条款。凡是注日期的引用文件，仅注日期的版本适用于本文件。凡是不注明日期的引用文件，其最新版本(包括所有的修改单)适用于本文件。

GB/T 6682 分析实验室用水规格和试验方法

3 方法提要

食品中增效醚残留用三氯甲烷提取，经氟罗里硅土柱净化，丙酮-三氯甲烷混合溶液洗脱，液相色谱-质谱质谱测定，外标法定量。

4 试剂和材料

除特别规定外，所用试剂均为分析纯，水为 GB/T 6682 规定的一级水。

4.1 乙腈：HPLC 级。

4.2 甲酸：HPLC 级，≥96%。

4.3 三氯甲烷。

4.4 正己烷。

4.5 丙酮。

4.6 氯化钠。

4.7 无水硫酸钠：使用前于 650 ℃灼烧 4 h，贮存干燥器中备用。

4.8 石墨化碳。

4.9 丙酮＋三氯甲烷(1＋2，体积比)。

4.10 0.1%甲酸溶液：量取 1 mL 甲酸用水定容至 1 000 mL。

4.11 乙腈＋0.1%甲酸溶液(80＋20，体积比)。

4.12 标准品：增效醚(piperonyl butoxide，CAS 号 51-03-6)，纯度≥94%。

4.13 标准储备溶液：准确称取增效醚标准品 10 mg(4.12)，用三氯甲烷溶解并定容至 100 mL，浓度相当于 100 μg/mL，储备液贮存于－18 ℃以下，有效期为 3 个月。

4.14 标准中间溶液：准确量取 1 mL 标准贮备溶液(4.13)，氮吹至干后用乙腈溶解并定容至 100 mL，该溶液浓度为 1 μg/mL，贮存于 2 ℃～4 ℃，有效期为 1 个月。

4.15 基质标准工作溶液：根据需要按本方法所述前处理步骤，制得空白样品提取液，用该样品提取液将标准中间溶液(4.14)稀释至适当浓度，该溶液使用前配制。

4.16 氟罗里硅土柱(florisil，1 g/3 mL)或相当者。

4.17 有机相滤膜：0.22 μm。

5 仪器和设备

5.1 液相色谱-质谱/质谱仪：配有电喷雾离子源(ESI)。

5.2 分析天平：感量 0.1 mg、0.01 g。

5.3 超声提取仪。

5.4 涡旋混匀器。

5.5 离心机：4 000 r/min。

5.6 固相萃取装置。

5.7 氮吹仪。

5.8 组织捣碎机。

5.9 粉碎机。

6 试样的制备与保存

6.1 猪肉、鸡肝和牛肾

从所取全部样品中取出有代表性样品可食部分约 500 g，用组织捣碎机充分捣碎均匀，装入洁净容器中，密封，并标明标记，于－18 ℃以下冷冻存放。

6.2 牛脂肪

从所取全部样品中取出有代表性样品约 500 g，于 50 ℃～60 ℃水浴中，使样品完全融化，充分混匀，装入洁净容器中，密封，并标明标记，于－18 ℃以下冷冻存放。

6.3 牛奶

从所取全部样品中取出有代表性样品约 500 g，充分混匀。装入洁净容器中，密封，并标明标记，于－18 ℃以下冷冻保存。

6.4 鸡蛋

从所取全部样品中取出有代表性样品约 500 g，去壳后混匀，装入洁净容器中，密封，并标明标记，于－18 ℃以下冷冻存放。

6.5 柑桔、大蒜、马铃薯、菠菜和芹菜

从所取全部样品中取出有代表性样品约 500 g，用组织捣碎机将样品加工成浆状，装入洁净容器中，密封，并标明标记，于－18 ℃以下冷冻保存。

6.6 大豆、花生、小麦、大米、大麦和芝麻

从所取全部样品取出有代表性样品 500 g，用粉碎机粉碎并通过 20# 目圆孔筛，混匀，装入洁净容器中，密封，并标明标记，于 2 ℃～4 ℃冷藏存放。

在制样过程中，应防止样品受到污染或发生残留物含量的变化。

7 测定步骤

7.1 提取

7.1.1 猪肉、鸡肝、牛肾、鸡蛋和牛奶

准确称取 1.0 g 均匀试样(精确至 0.01 g)于 50 mL 离心管中,加入 1 g 无水硫酸钠(4.7)、0.5 g 氯化钠,混合均匀后,加入 12.5 mL 三氯甲烷,涡旋混匀 3 min,超声提取 10 min,4 000 r/min 离心 5 min,转移上清液至另一具刻度试管。再加入 12.5 mL 三氯甲烷,重复涡旋混匀、超声、离心步骤,合并上清液,并定容至 25 mL。

7.1.2 牛脂肪

准确称取 1.0 g 均匀试样(精确至 0.01 g)于 50 mL 离心管中,将样品置于 50 ℃～60 ℃水浴中加热,待样品完全融化后取出,加入 0.5 g 氯化钠,并用三氯甲烷溶解至 25 mL,涡旋混匀 3 min,超声提取 10 min,4 000 r/min 离心 5 min,转移上清液至另一试管。

7.1.3 柑桔、大蒜、马铃薯、菠菜和芹菜

准确称取 1.0 g 均匀试样(精确至 0.01 g)于 50 mL 离心管中,加入 1 g 无水硫酸钠(4.7)、0.5 g 氯化钠、0.1 g 石墨化碳、12.5 mL 三氯甲烷,涡旋混匀 3 min,超声提取 10 min,4 000 r/min 离心 5 min,转移上清液至另一具刻度试管。再加入 12.5 mL 三氯甲烷,重复涡旋混匀、超声、离心步骤,合并上清液,并定容至 25 mL。

7.1.4 大豆、花生、小麦、大米、大麦和芝麻

准确称取 1.0 g 均匀试样(精确至 0.01 g)于 50 mL 离心管中,加入 0.5 g 氯化钠、12.5 mL 三氯甲烷,涡旋混匀 3 min,超声提取 10 min,4 000 r/min 离心 5 min,转移上清液至另一具刻度试管。再加入 12.5 mL 三氯甲烷,重复涡旋混匀、超声、离心步骤,合并上清液,并定容至 25 mL。

7.2 净化

7.2.1 牛脂肪

加入 3 mL 三氯甲烷活化氟罗里硅土柱(4.16),取 5 mL 样液以小于 1 mL/min 的流速通过柱体,待样液全部流过后,加入 5 mL 正己烷、5 mL 三氯甲烷淋洗,抽干柱体后,用 6 mL 丙酮＋三氯甲烷(4.9)洗脱固相萃取柱,收集洗脱液于 50 ℃水浴中氮吹浓缩至干,加入 1 mL 乙腈＋0.1%甲酸溶液(4.11)溶解残渣,过 0.22 μm 滤膜,待测定。

7.2.2 其他食品(除牛脂肪外)

加入 3 mL 三氯甲烷活化氟罗里硅土柱(4.16),将 5 mL 上清液样液以小于 1 mL/min 的流速通过柱体,待样液全部流过后,加入 5 mL 三氯甲烷淋洗,抽干柱体后,用 6 mL 丙酮＋三氯甲烷(4.9)洗脱固相萃取柱,收集洗脱液于 50 ℃水浴中氮吹浓缩至干,加入 1 mL 乙腈＋0.1%甲酸(4.11)溶解残渣,过 0.22 μm 滤膜,待测定。

7.3 测定

7.3.1 液相色谱参考条件

液相色谱参考条件如下:

a) 色谱柱：Zobax SB C_{18}，150×2.1 mm(i.d.)，5 μm 或相当者；
b) 流动相：乙腈+0.1%甲酸溶液，(80+20，体积比)；
c) 流速：0.3 mL/min；
d) 柱温：30 ℃；
e) 进样量：10 μL。

7.3.2 质谱条件

质谱条件如下：
a) 离子化模式：电喷雾电离正离子模式(ESI^+)；
b) 质谱扫描方式：多反应监测(MRM)；
c) 定性离子对：356.3/177.2，356.3/194.1；
d) 定量离子对：356.3/177.2；
e) 其他质谱参考条件参见附录 A。

7.3.3 定量测定

根据样液中被测增效醚残留量的情况，选定峰面积相近的标准工作溶液。标准工作溶液和样液中增效醚残留的响应值均应在仪器的检测线性范围内。对标准工作溶液和样液等体积参差进样测定。增效醚参考保留时间约为 4.0 min；标准溶液的液相色谱-质谱/质谱多反应监测(MRM)色谱图参见图 B.1。

7.3.4 定性测定

按照上述仪器条件测定样品和标准工作溶液，如果样品的质量色谱峰相对保留时间与标准溶液在±2.5%范围内；定性离子对的相对丰度与浓度相当的标准溶液的相对丰度一致，相对丰度偏差不超过表 1 的规定，则可判断样品中存在相应的被测物。

表 1 定性测定时相对离子丰度的最大允许偏差

相对离子丰度/%	>50	>20～50	>10～20	≤10
允许的相对偏差/%	±20	±25	±30	±50

8 空白试验

除不加试样外，均按上述测定步骤进行。

9 结果计算与表述

9.1 采用外标法定量，按式(1)计算增效醚残留量：

$$X_i = \frac{A_i \cdot c_s \cdot V}{A_s \cdot m} \quad \cdots\cdots(1)$$

式中：
X_i ——试样中增效醚的残留量，单位为微克每千克(μg/kg)；
A_i ——样液中增效醚的色谱峰面积；
c_s ——标准工作液中增效醚的浓度，单位为微克每升(μg/L)；

V ——样液最终定容体积，单位为毫升(mL)；

A_s ——标准工作液中增效醚的色谱峰面积；

m ——最终样液所代表的试样量，单位为克(g)。

9.2 计算结果应扣除空白值。

10 测定低限与回收率

10.1 测定低限

本方法的测定低限为 10 μg/kg。

10.2 回收率

回收率数据见表 2。

表 2 方法回收率数据

食品	添加水平 μg/kg	回收率 %	食品	添加水平 μg/kg	回收率 %
柑桔	10	72.0～102.0	猪肉	10	70.0～90.0
	200	78.4～96.6		80	79.2～94.4
	8 000	85.0～97.2		7 000	88.8～102.1
花生	10	76.0～92.0	鸡肝	10	72.0～90.0
	50	76.5～92.0		1 000	77.4～96.6
	8 000	82.6～94.4		10 000	84.0～99.7
大蒜	10	72.0～96.0	牛肾	10	74.0～104.0
	500	73.4～100.2		200	71.6～93.0
	8 000	79.0～96.1		10 000	84.7～99.4
土豆	10	70.0～98.0	牛脂肪	10	66.0～92.0
	300	76.0～93.0		30	67.2～87.2
	8 000	84.2～94.2		7 000	77.9～94.6
菠菜	10	70.0～90.0	鸡蛋	10	72.0～96.0
	300	81.2～100.4		20	77.0～96.6
	50 000	82.7～100.5		1 000	87.8～102.7
芹菜	10	66.0～94.0	牛奶	10	66.0～96.0
	500	84.0～93.4		50	74.0～92.0
	8 000	83.2～100.8		200	79.5～96.0
小麦	10	70.0～86.0	芝麻籽	10	65.2～113.0
	8 000	72.5～91.0		500	72.3～100.1
	30 000	75.8～91.4		80 000	86.4～99.2
大豆	10	67.1～106.3	大米	10	82.2～105.4
	200	74.7～91.4		20 000	82.4～96.3
	8 000	79.2～98.5		30 000	87.0～95.4
大麦	10	81.0～102.9	—	—	
	20 000	78.9～91.7			
	30 000	84.8～98.8			

附 录 A
（资料性附录）
参考质谱参数

表 A.1 参考质谱参数

质谱参数	参数值
雾化气/psi	25
气帘气/psi	25
辅助加热气/psi	20
碰撞/psi	12
源温度/℃	400
喷雾电压/V	4 000
射入电压/V	10
碰撞室出口电压/V	12
去簇电压/V	40(356.30/177.20)、45(356.30/194.10)
碰撞能量/eV	16(356.30/177.20)、11(356.30/194.10)

非商业性声明：附录 A 所列参数是用 API4000 质谱仪完成的，此处列出试验用仪器型号仅是为了提供参考，并不涉及商业目的，鼓励标准使用者尝试采用不同厂家或型号的仪器。

附 录 B
（资料性附录）
增效醚标准溶液多反应监测（MRM）色谱图

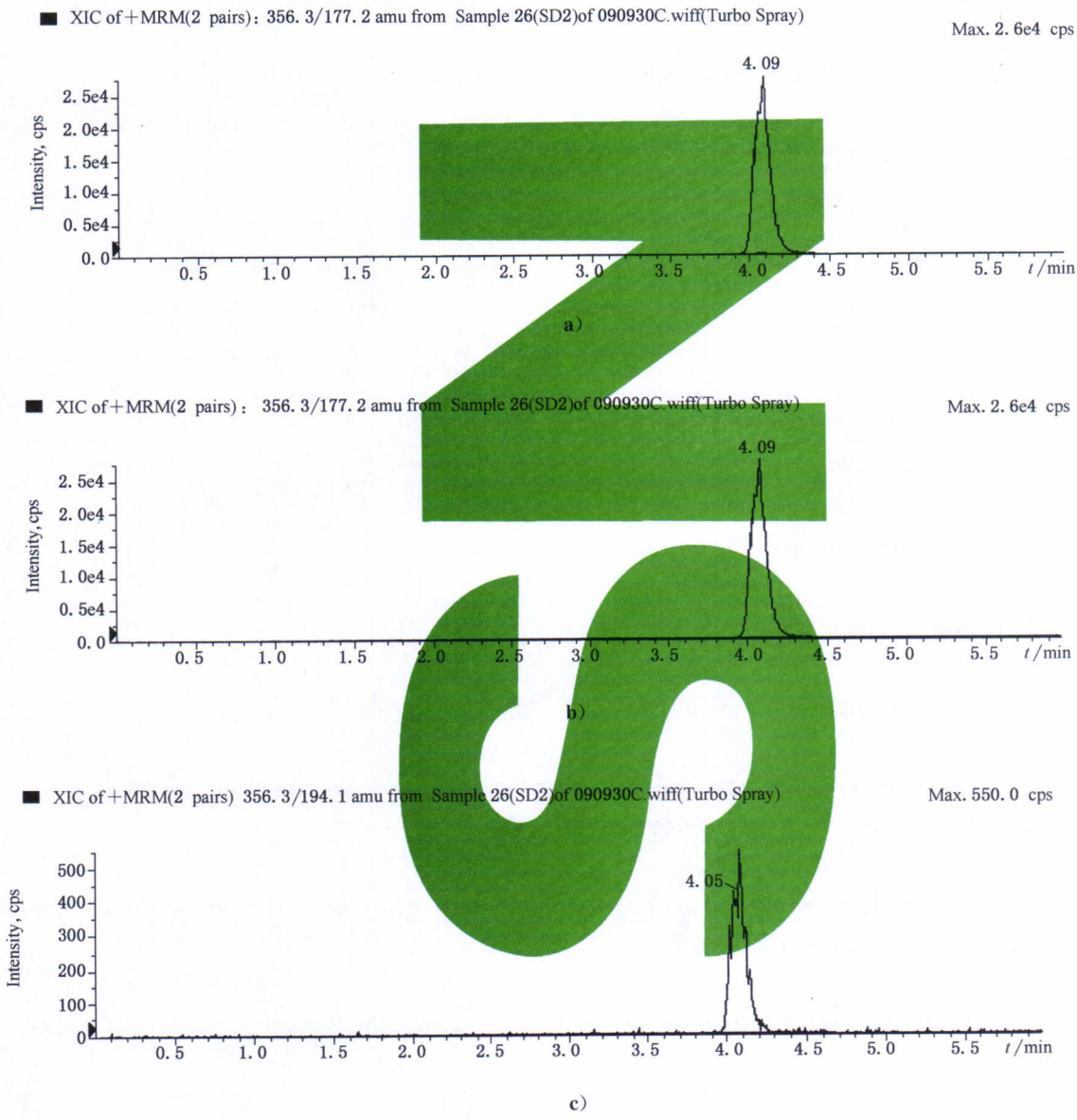

图 B.1 增效醚标准溶液（2 ng/mL）多反应监测（MRM）色谱图

Foreword

The standard replaced SN 0526—1996 Method for the determination of piperonyl butoxide residues in cereals for export.

Compared to SN 0526—1996, the main changes are as follows:

—Changed the standard name

—Deleted the"sampling"part

—Changed the preparation method

—Changed the detection method

—Increased the sample matices

—Changed the standard structure

—Reduced the limit of quantification

This standards is drafted accordance with GB/T 1.1—2009 given rules.

Please note that some of the contents of this standard may involve patents. This standars publisher have not responsibility for identifying these patents.

This standard is proposed by and is under the charge of the Certification and Accreditation administration of the People's Republic of China.

This standard is drafted by Henan Entry-Exit Inspection and Quarantine Bureau of the People's Republic of China.

The main drafters of this standard are yang jizhou, zhu weixia, yuan ping, liu yafeng, sun zhuanlian, wei wei, wang caijuan.

Note: This English version is a translation from the Chinese text, is solely for guidance.

Determination of piperonyl butoxide residues in foodstuffs for export—LC-MS/MS method

1 Scope

The standard specifies and the method of sampling and determination of piperonyl butoxide residue in foodstuffs for export by LC-MS/MS method.

The method is applicable to determine piperonyl butoxide residue in pork, chicken liver, bovine kidney, bovine fat, milk, egg, orange, bean, peanut, garlic, potato spinach, celery, wheat, sesame, rice and barley for export by LC-MS/MS method.

2 Normative references

The following documents is necessary for this standard. For dated references, only dated editions shall apply to this standard. For undated references, the last edition of the normative document (including subsequent amendments) reffered to applies.

GB/T 6682 water for analytical laboratory use—specification and test methods

3 Principle

Extraction of the piperonyl butoxide residue with chloroform. The purification procedures were with a florisil solid phase extraction (SPE) cartridge. Actone-chloroform is used eluted solvent. The piperonyl butoxide residues were determined by liquid chromatography-tandem mass spectrometry and quantified with external standard method.

4 Reagents and Materials

Unless otherwise specified, all regents used are A.R. grade, and water is the first grade according to GB/T 6682.

4.1 Acetonitrile: HPLC grade.

4.2 Formic acid: HPLC grade, ≥96%.

4.3 Chloroform.

4.4 n-Hexan.

4.5 Acetone.

4.6 Sodium Chloride.

4.7 Sodium sulfate: burning for 4 h before using and storing in desiccator.

4.8 Graphitized carbon.

4.9 Acetone + chloroform(1 + 2, *V*/*V*).

4.10 0.1% formic acid: 1 mL formic acid diluted to 1 000 mL with water.

4.11 Acetonitrile + 0.1% formic acid(80 + 20, *V*/*V*).

4.12 Standard of piperonyl butoxide: Piperonyl butoxide (CAS No.51-03-6), purity≥94%.

4.13 Stock solutions of piperonyl butoxide: Accurately weigh piperonyl butoxide standard material 10 mg (4.12), dissolve with chloroform to a volume of 100 mL, the soltuion concentration is 100 μg/mL and store below −18 ℃ for a maximum period of 3 months.

4.14 Middle standard solution: taken out 1 mL stock standar solution (4.13) and evaporated to dryness. Added acetonitrile to dissolve and to 100 mL. The concentration of the middle solution is 1 μg/mL. The solution should be stored at the temperature 2 ℃～4 ℃ and is stable for one month.

4.15 Working standard solution: used the described method and the blank matrix solution were obtained. Dilute appropriate volume of middle standard solutions(4.14) to a intended concentration with the matix solution, and prepare freshly.

4.16 florisil solid phase extraction catridge: 1 g/3 mL or equivalent.

4.17 Filter membrane: 0.22 μm, organic.

5 Apparatus

5.1 Liquid chromatography-tandem mass spectrometry: Equipped with electrospray (ESI) ion source.

5.2 Electronic balance: Accuracy 0.1 mg and 0.01 g, respectively.

5.3 Ultrasonic machine.

5.4 Vortex mixer.

5.5 Centrifuge: 4 000 r/min.

5.6 Solid phase extraction equipment.

5.7 Nitrogen gas blowing concentrator.

5.8 Tissues homogenizer.

5.9 Grinder.

6 Sample preparation and storage

6.1 Pork, chicken liver and bovine kidney tissues.

Collect 500 g the representative samples and the edible portions are mixed well with a tissure homogenizer, seal in two clean containers and label. The sample was stored at below −18 ℃.

6.2 Bovine fat.

Collect 500 g the representative samples and melted with water bath under 50 ℃～60 ℃. After mixing, seal in two clean containers and label. The sample was stored at below −18 ℃.

6.3 Milk.

Collect 500 g the representative samples. After mixing, seal in two clean containers and label. The sample was stored at below −18 ℃.

6.4 Egg.

Collect 500 g the representative samples and the edible portions are mixed, seal in two clean containers and label. The sample was stored at below −18 ℃.

6.5 Orange, garlic, potato spinach and celery.

Collect 500 g the representative samples the edible portions are cut up and mixed well with a tissure homogenizer, divide the prepared samples, seal in two clean containers and label. The sample was stored at below −18 ℃.

6.6 Bean, peanut, wheat, sesame, rice and barley.

Collect 500 g the representative samples and crush with a grinder, let them pass through a 20 mesh sieve, divide the prepared samples, eal in two clean containers and label. The sample was stored at 2 ℃～4 ℃.

In the course of sample preparation, precautions must be taken to avoid contamination or any factors, which may cause the change of residue content.

7 Method of Determination

7.1 Extraction

7.1.1 Pork, chicken liver, bovine kidney, eggs, milk.

Weigh 1.0 g of the prepared test samples into a 50 mL centrifuge tube(accurated to 0.01 g), adding 1 g sodium sulfate(4.7) and 0.5 g sodium chloride. After mixing well, then adding 12.5 mL chroloform, extract with vortex mixer for 3 min and for ultersonic machine for 10 min, centrifuge for 5 min at 4 000 r/min and then, transfer the supernatant to another clean tube. Repeat the extraction procedure again using 12.5 mL chloroform. Combined the entire supernatants and volume to 25 mL with chloroform.

7.1.2 bovine fat.

Weigh 1.0 g of the prepared test samples into a 50 mL centrifuge tube(accurated to 0.01 g). the sample was melted with water bath under 50 ℃～60 ℃. Afer taken out, adding 0.5 g sodium chloride and 25 mL chroloform, extract with vortex mixer for 3 min and for ultersonic machine for 10 min, centrifuge for 5 min at 4 000 r/min and then, transfer the supernatant to another clean tube.

7.1.3 Orange, garlic, potato spinach and celery.

Weigh 1.0 g of the prepared test samples into a 50 mL centrifuge tube(accurated to 0.01 g),adding 1 g sodium sulfate(4.7), 0.5 g sodium chloride and 0.1 graphitized carbon. After mixing well, then adding 12.5 mL chroloform, extract with vortex mixer for 3 min and for ultersonic machine for 10 min, centrifuge for 5 min at 4 000 r/min and then, transfer the supernatant to another clean tube. Repeat the extraction procedure again using 12.5 mL chloroform. Combined the entire supernatants and volume to 25 mL with chloroform.

7.1.4 Bean, peanut, wheat, sesame, rice and barley.

Weigh 1.0 g of the prepared test samples into a 50 mL centrifuge tube(accurated to 0.01 g),adding 0.5 g sodium chloride and 12.5 mL chroloform, extract with vortex mixer for 3 min and for ultersonic machine for 10 min, centrifuge for 5 min at 4 000 r/min and then, transfer the supernatant to another clean tube.Repeat the extraction procedure again using 12.5 mL chloroform.Combined the entire supernatants and volume to 25 mL with chloroform.

7.2 Clean-up

7.2.1 Bovine fat.

Condition a fiorisil SPE column(4.16) with 3 mL chloroform, Allow the 5 mL extract to permeate at less than the speed of 1 mL/min through the cartridge. After penetration, the column is washed successively with 5 mL n-hexan and 5 mL chloroform dried the column by sucking air. The analyte is eluted with 6 mL acetone-chloroform(4.9). The eluate is evaporated to dryness under a stream of nitrogen at 50 ℃ and the residues were dissolved with acetonitrile + 0.1% formic acid (4.11). After filtered with 0.22 μm filter, the final solution is ready for LC-MS/MS determination.

7.2.2 All the foodstuffs, except bovine fat

Condition a florisil SPE column(4.16) with 3 mL chloroform, Allow the 5 mL extract to permeate at less than the speed of 1 mL/min through the cartridge. After penetration, the column is washed successively with 5 mL chloroform dried the column by sucking air. The analyte is eluted with 6 mL acetone-chloroform(4.9). The eluate is evaporated to dryness under a stream of nitrogen at 50 ℃ and the residues were dissolved with acetonitrile + 0.1% formic acid (4.11). After filtered with 0.22 μm filter, the final solution is ready for LC-MS/MS determination.

7.3 Determination

7.3.1 LC operation conditions

LC operation conditions are as follows:

a) Column: Zobax SB C_{18} column, 150 mm × 2.1 mm(i.d.), 5 μm, or equivalent.

b) Mobile phase: acetonitrile + 0.1% formic acid (80 + 20, *V*/*V*).

c) Flow rate: 0.3 mL/min.

d) Column temperature: 30 ℃.

e) Injection volume: 10 μL.

7.3.2 MS operation conditions

MS operation conditions are as follows:

a) Ion mode: ESI, positive ionisation mode.

b) Scan mode: multiple reaction monitoring(MRM) mode.

c) Qulitication trantition:356.3/177.2, 356.3/194.1.

d) Quantification trantition: 356.3/177.2.

e) Other reference mass operating conditions are listed in Talbe A.1 in Annex A.

7.3.3 LC-MS detection

Prepare standard solutions containing piperonyl butoxide at appropriate concentrations according to the analyte in sample extracts. The referenced retention times for piperonyl butoxide are 4.0 min. Annex B are the reconstituted ion chromatograms of piperonyl butoxide standard solution.

7.3.4 Confirmation test

The qualification ions of the analyte must be found, and at least include one precursor ion and two daughter ions. For the same analysis batch and the same analyte, the variation range of the ion ratio

between the two daughter ions for the unknown samples and the standard working solutions at the similar concentration can not be out of range of table 1 under the same determination conditions.

Table 1—Maximum permitted tolerances for relative ion intensities

Relative intensity(% of base peak)	>50	>20~50	>10~20	≤10
Maximum permitted tolerances for relative ion intensities/%	±20	±25	±30	±50

8 Blank test

The operation of the blank test is the same as that described in the method of determination, but with omission of sample addition.

9 Calculation and expression of the result

9.1 Quantify by external standard method. Calculate the concentration of the piperonyl butoxide residue in sample according to equation (1):

$$X_i = \frac{A_i \cdot c_s \cdot V}{A_s \cdot m} \quad \cdots\cdots (1)$$

Where:

X_i —the residue content of piperonyl butoxide in the test sample, (μg/kg);

A_i —the corresponding area of piperonyl butoxide in the test solution;

c_s —the concentration of piperonyl butoxide in the matrix standard solution, (μg/L);

V —the final volume of sample solution, (mL);

A_s —the corresponding area of piperonyl butoxide in the matrix standard solution;

m —the corresponding mass of test sample in the final sample solution, (g).

9.2 The results should be deduct blank value.

10 Limit of quantification and recovery

10.1 Limit of quantification

The limit of quantification of piperonyl butoxide is 10 μg/kg.

10.2 Recovery

The recovery data of piperonyl butoxide residues determined by HPLC-MS/MS is listed in table 2.

Table 2—Recovery for piperonyl butoxide residue

Matrix	Spiked level μg/kg	Recovery %	Matrix	Spiked level μg/kg	Recovery %
orange	10	72.0～102.0	pork	10	70.0～90.0
	200	78.4～96.6		80	79.2～94.4
	8 000	85.0～97.2		7 000	88.8～102.1
peanut	10	76.0～92.0	liver	10	72.0～90.0
	50	76.5～92.0		1 000	77.4～96.6
	8 000	82.6～94.4		10 000	84.0～99.7
garlic	10	72.0～96.0	kidney	10	74.0～104.0
	500	73.4～100.2		200	71.6～93.0
	8 000	79.0～96.1		10 000	84.7～99.4
potato	10	70.0～98.0	fat	10	66.0～92.0
	300	76.0～93.0		30	67.2～87.2
	8 000	84.2～94.2		7 000	77.9～94.6
spinach	10	70.0～90.0	egg	10	72.0～96.0
	300	81.2～100.4		20	77.0～96.6
	50 000	82.7～100.5		1 000	87.8～102.7
celery	10	66.0～94.0	milk	10	66.0～96.0
	500	84.0～93.4		50	74.0～92.0
	8 000	83.2～100.8		200	79.5～96.0
wheat	10	70.0～86.0	sesame	10	65.2～113.0
	8 000	72.5～91.0		500	72.3～100.1
	30 000	75.8～91.4		80 000	86.4～99.2
bean	10	67.1～106.3	rice	10	82.2～105.4
	200	74.7～91.4		20 000	82.4～96.3
	8 000	79.2～98.5		30 000	87.0～95.4
barley	10	81.0～102.9	—	—	
	20 000	78.9～91.7			
	30 000	84.8～98.8			

Annex A
(Informative)
Reference mass conditions

Table A.1—Main Mass parameters of piperonyl butoxide

Mass parameter	Values
Gas 1/psi	25
Curtain gas/psi	25
Gas 2/psi	20
CAD/psi	12
Temperature/℃	400
IS/V	4 000
EP/V	10
CXP/V	12
DP/V	40(356.30/177.20)、45(356.30/194.10)
CE/eV	16(356.30/177.20)、11(356.30/194.10)

Declaration for non-commercial: The reference mass conditions listed in Annex A are performed on API 4 000 mass spectrum. The type of the equipment mentioned here is only for reference and not for commercial purpose. Encourage users to try different manufactures or models of equipments.

Annex B
(Informative Annex)
MRM chromatogram of piperonyl butoxide standard solution

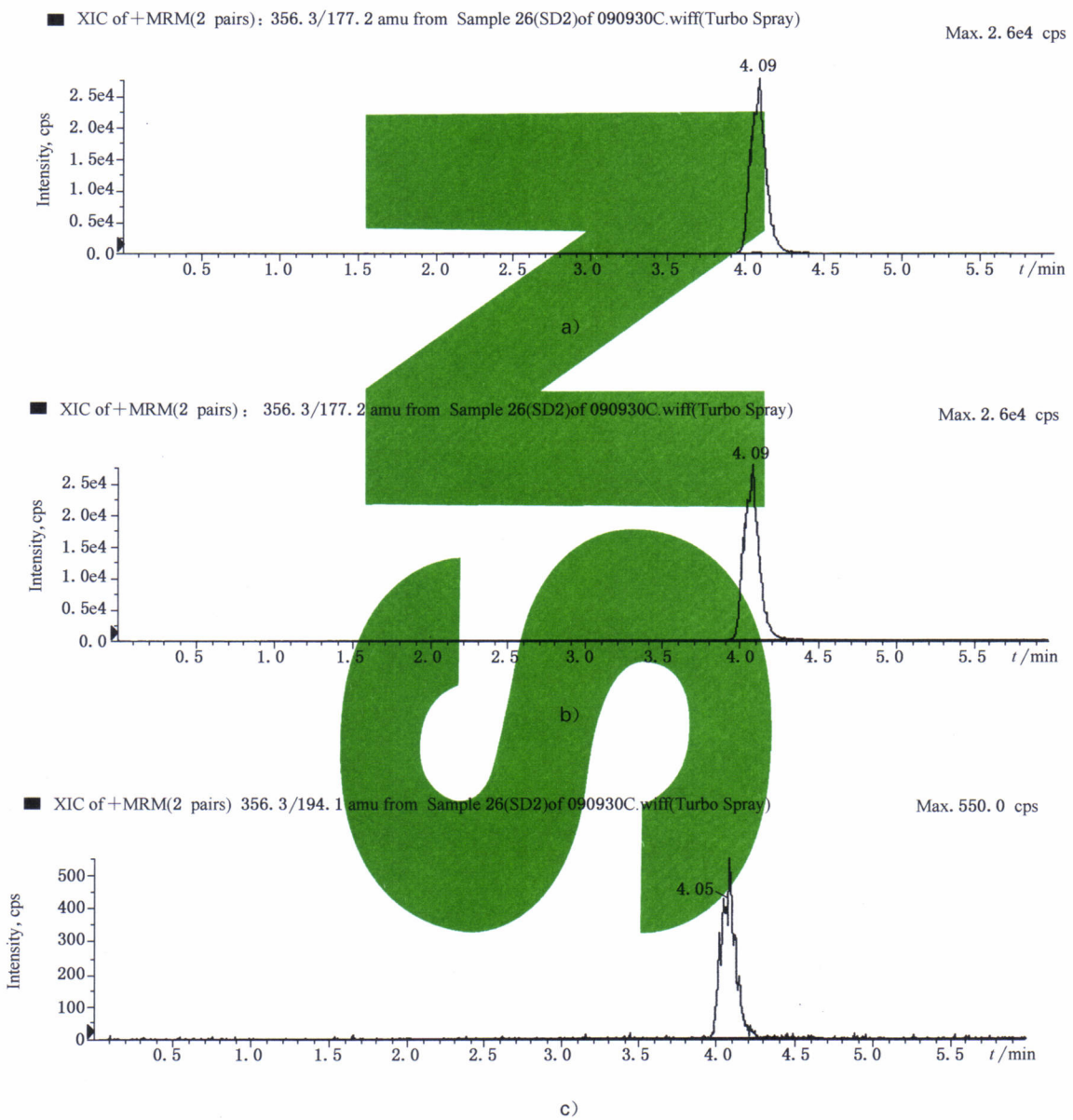

Fig.B.1—MRM chromatogram of piperonyl butoxide standard solution(2 ng/mL)