SN

中华人民共和国出入境检验检疫行业标准

SN/T 0639—2013
代替 SN 0639—1997

出口肉及肉制品中利谷隆及其代谢产物残留量的检测方法 液相色谱-质谱/质谱法

Determination of linuron and its metabolite residues in meat and meat products for export— HPLC-MS/MS method

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前　言

本标准按照 GB/T 1.1—2009 给出的规则起草。

本标准代替 SN 0639—1997《出口肉及肉制品中利谷隆残留量检验方法》。

本标准与 SN 0639—1997 相比，主要技术变化如下：

——更改了标准名称，增加了代谢物的测定；

——扩大了适用的基质范围；

——删除了抽样部分；

——气相色谱法更改为液相色谱-质谱/质谱法，降低了方法的检出限。

请注意本文件的某些内容可能涉及专利。本文件的发布机构不承担识别这些专利的责任。

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：中华人民共和国上海出入境检验检疫局。

本标准主要起草人：伊雄海、曲栗、曹晨、郭德华、邓晓军、朱坚、盛永刚、杨惠琴、包明。

本标准所代替标准的历次版本发布情况为：

——SN 0693—1997。

出口肉及肉制品中利谷隆及其代谢产物残留量的检测方法 液相色谱-质谱/质谱法

1 范围

本标准规定了出口肉和肉制品中利谷隆及其代谢物(3,4-二氯苯胺)残留量的测定方法。

本标准适用于出口猪肉、猪肾、猪肝、猪肠衣、午餐肉、香肠等肉及肉制品中利谷隆及其代谢物(3,4-二氯苯胺)残留量的液相色谱-质谱/质谱测定和确证。

2 规范性引用文件

下列文件对于本文件的应用是必不可少的。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

试样中残留的利谷隆及其代谢产物用丙酮-乙腈提取,提取液于-18 ℃冰箱中冷冻去脂,提取液采用弗罗里硅土柱净化,液相色谱-质谱/质谱检测和确证,内标法定量。

4 试剂与材料

除非另有规定,均使用分析纯试剂,水为去离子水。

4.1 乙腈:高效液相色谱级。

4.2 丙酮:高效液相色谱级。

4.3 正己烷:高效液相色谱级。

4.4 乙醚:高效液相色谱级。

4.5 甲酸:优级纯,≥99%。

4.6 乙酸铵。

4.7 丙酮+乙腈(5+95,体积比):量取 50 mL 丙酮和 950 mL 乙腈,混匀。

4.8 乙醚+正己烷(1+9:体积比):量取 100 mL 乙醚和 900 mL 正己烷,混匀。

4.9 丙酮+正己烷(1+9:体积比):量取 100 mL 丙酮和 900 mL 正己烷,混匀。

4.10 乙腈+水(1+1,体积比):量取 100 mL 乙腈和 100 mL 水,混匀。

4.11 无水硫酸钠:650 ℃灼烧 4 h,贮于密封容器中备用。

4.12 含 5 mmol/L 乙酸铵和 0.1%甲酸的水溶液:精确称取 385.4 mg 乙酸铵和 1 mL 甲酸溶解于纯水中并定容至 1 L。

4.13 标准物质:利谷隆(Linuron,CAS 号:330-55-2)、利谷隆同位素内标(Linuron-D6)、3,4-二氯苯胺(3,4-Dichloroaniline,CAS 号:95-76-1);3,4-二氯苯胺同位素内标(3,4-Dichloroaniline-2,6-D2),纯度

均大于98%。

4.14 标准储备液的配制：分别精确称取利谷隆、利谷隆同位素内标、3,4-二氯苯胺以及3,4-二氯苯胺同位素内标的标准品0.01 g(精确到0.000 1 g)，用乙腈溶解，稀释并定容于10 mL容量瓶中，分别配制成浓度为1 mg/mL的标准储备溶液，−18 ℃以下避光保存，有效期12个月。

4.15 标准中间储备溶液的配制：准确移取100 μL上述各标准储备液于100 mL容量瓶中，用乙腈稀释定容，分别配成1 μg/mL的利谷隆、利谷隆同位素内标、3,4-二氯苯胺以及3,4-二氯苯胺同位素内标标准溶液，−18 ℃以下避光保存，有效期6个月。

4.16 基质标准工作溶液的配制：准确移取适量上述标准中间储备溶液，用基质空白溶液配制成适当浓度的混合标准工作曲线溶液，于4 ℃避光冷藏保存，临用时现配。

4.17 微孔滤膜：0.45 μm，有机系。

4.18 弗罗里硅土小柱：6 mL，1 g或相当者。使用前用5 mL正己烷过柱活化。

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0.000 1 g和0.01 g。

5.3 涡旋混匀器。

5.4 冷冻离心机：10 000 r/min。

5.5 固相萃取装置。

5.6 氮吹仪。

5.7 −20 ℃冰箱。

5.8 离心管：50 mL。

5.9 玻璃试管：25 mL，具刻度。

6 试样制备与保存

6.1 试样制备

从所取全部试样中取出有代表性试样可食部分约500 g，充分捣碎均匀，装入洁净容器中，密封，并标明标记。肠衣试样在提取前先用去离子水洗，去盐。

6.2 试样保存

于−18 ℃以下冷冻存放。在制样的操作过程中，应防止样品污染或发生残留物含量的变化。

7 分析步骤

7.1 提取

称取均质试样2 g(精确至0.01 g)，置于50 mL塑料离心管中，分别加入100 μL的利谷隆内标溶液和3,4-二氯苯胺内标溶液(4.15)，加入2 g无水硫酸钠，研磨均匀，加入15 mL丙酮+乙腈(5+95，体积比)(4.7)，匀质2 min，以3 000 r/min离心3 min，转移上层有机相，收集于另一50 mL塑料离心管中，残渣再用10 mL丙酮+乙腈(5+95，体积比)按上述步骤提取一次，合并提取液，待净化。

7.2 净化

将提取液于−18 ℃冰箱中静置1 h，然后迅速过滤于具刻度玻璃试管，用丙酮定容至25 mL，取

5 mL 样品于 15 mL 玻璃试管中，于室温下氮吹至近干，加入 2 mL 正己烷振荡溶解。

将上述所得样液转移至弗罗里硅土柱中，并用 2 mL 乙醚＋正己烷(1＋9，体积比)(4.8)洗涤 15 mL 离心管，倒入柱内，用 4 mL 乙醚＋正己烷(1＋9，体积比)淋洗小柱，弃去淋出液，抽干小柱。用 7.5 mL 丙酮＋正己烷(1＋9，体积比)(4.9)分 3 次洗脱固相萃取柱上待分析组分，流速约 30 滴/min，收集洗脱液，在 40 ℃条件下氮吹至近干，加入 1.0 mL 乙腈＋水(1＋1，体积比)(4.10)振荡溶解残渣，过 0.45 μm 有机型滤膜后，液相色谱-质谱仪进行测定。

7.3 测定

7.3.1 高效液相色谱条件

高效液相色谱条件如下：

a) 色谱柱：C_{18}色谱柱，50 mm×4.6 mm(内径)，粒径 1.8 μm，或相当者；

b) 流动相：A：5 mmol/L 乙酸铵＋0.1%甲酸溶液；B：乙腈，梯度洗脱程序见表 1；

c) 流速：0.3 mL/min；

d) 进样量：20 μL；

e) 柱温：室温。

表 1 流动相及梯度洗脱条件

时间/min	乙腈/%	5 mmol/L 乙酸铵＋0.1%甲酸溶液/%
0.0	2	98
2.0	2	98
5.5	99	1
11.1	99	1
11.2	2	98
18.0	2	98

7.3.2 质谱条件

质谱条件如下：

a) 离子源：电喷雾 ESI，正模式；

b) 扫描方式：多反应监测 MRM；

c) 雾化气(GS1)、气帘气(CUR)、辅助气(GS2)、碰撞气(CAD)均为高纯氮气或其他合适气体；使用前应调解各气体流量以及离子源温度(TEM)使质谱灵敏度达到检测要求，详细条件参见附录 A；

d) 电喷雾电压(IS)、碰撞电压(CE)、去簇电压(DP)、碰撞室入口电压(EP)、碰撞室出口电压(CXP)应优化至最佳灵敏度，监测离子和定量离子等详细条件参见附录 A 中表 A.1。

7.3.3 定量测定

根据试样中被测物的含量情况，选取响应值适宜的标准工作液进行色谱分析，标准曲线工作液应有 5 个浓度水平。标准工作液和待测液中两种药物的响应值均应在仪器线性响应范围内。如果样品中待测物含量超过线性范围，应用流动相稀释到合适浓度后分析。在上述色谱条件下的利谷隆和 3，4-二氯苯胺的参考保留时间为分别为 8.5 min、8.0 min 左右，利谷隆和 3，4-二氯苯胺标准溶液的多反应监测

(MRM)色谱图参见附录B图B.1。

7.3.4 定性测定

按照液相色谱-质谱/质谱条件测定试样和标准工作溶液，如果试样中待测物质的保留时间与标准品一致，保留时间偏差在5%之内；定性离子对的相对丰度，是用相对于最强离子丰度的强度百分比表示，应当与浓度相当标准工作溶液的相对丰度一致，相对丰度允许偏差不超过表2规定的范围，则可判定为试样中存在对应的待测物。

表2 定性确证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

7.4 空白试验

除不加试样外，均按上述操作步骤进行。

8 结果计算和表述

用色谱数据处理机或按式(1)计算试样中药物的残留含量，计算结果需扣除空白值：

$$X_i = \frac{R_i \times c_i \times V}{R_s \times m} \times \frac{1\,000}{1\,000} \qquad \cdots\cdots(1)$$

式中：

X_i ——试样中利谷隆或3,4-二氯苯胺残留量，单位为微克每千克(μg/kg)；

R_i ——样液中的分析物与内标物峰面积的比值；

c_i ——从标准曲线上得到的药物残留量的溶液浓度，单位为纳克每毫升(ng/mL)；

V ——样液最终定容体积，单位为毫升(mL)；

R_s ——标准曲线中分析物与内标物峰面积的比值；

m ——最终样液代表的试样质量，单位为克(g)。

9 方法的测定低限和回收率

9.1 测定低限(LOQ)

本方法对利谷隆和或3,4-二氯苯胺的测定低限为10 μg/kg。

9.2 回收率

采用本方法对猪肉、猪肾、猪肝、猪肠衣、午餐肉和香肠食品基质进行添加回收试验，两种待测物在各个基质中的回收率资料参见附录C中表C.1。

附 录 A
（资料性附录）
API 4000 四级杆质谱仪参数[1)]

质谱仪参数：

a) 电喷雾电压(IS)：正模式 5 500 V；

b) 碰撞气压力(CAD)：Medium；

c) 雾化气压力(GS1)：517 kPa(70 psi)；

d) 气帘气压力(CUR)：172 kPa(30 psi)；

e) 辅助气流速(GS2)：414 kPa(60 psi)；

f) 离子源温度(TEM)：550 ℃；

g) 监测离子对、碰撞电压(CE)、去簇电压(DP)、碰撞室入口电压(EP)、碰撞室出口电压(CXP)如表 A.1 所示。

表 A.1 利谷隆和 3,4-二氯苯胺定性、定量离子对以及 CE、DP、EP、CXP 参考值

化合物	母离子(Q1)	子离子(Q3)	CE(V)	DP(V)	EP(V)	CXP(V)
利谷隆	249.1	160.1*	24.5	60.0	9.0	13.0
		182.0	22.0	60.0	9.0	11.0
D6-利谷隆同位素内标	255.1	160.1*	23.0	64.0	12.0	9.0
3,4-二氯苯胺	162.0	109.1	44.0	67.0	8.0	21.0
		127.1*	28.0	67.0	8.0	25.0
D2-3,4-二氯苯胺同位素内标	164.1	129.1*	28.5	60.0	6.0	25.0
注：* 为定量离子对。						

1) 非商业性声明：附录 A 所列参考质谱条件是在 AB API 4000 Q TRAP 质谱仪上完成的，此处列出试验用仪器型号仅为提供参考，并不涉及商业目的，鼓励标准使用者尝试不同厂家或型号的仪器。

附 录 B
（资料性附录）
利谷隆和 3,4-二氯苯胺标准品色谱图

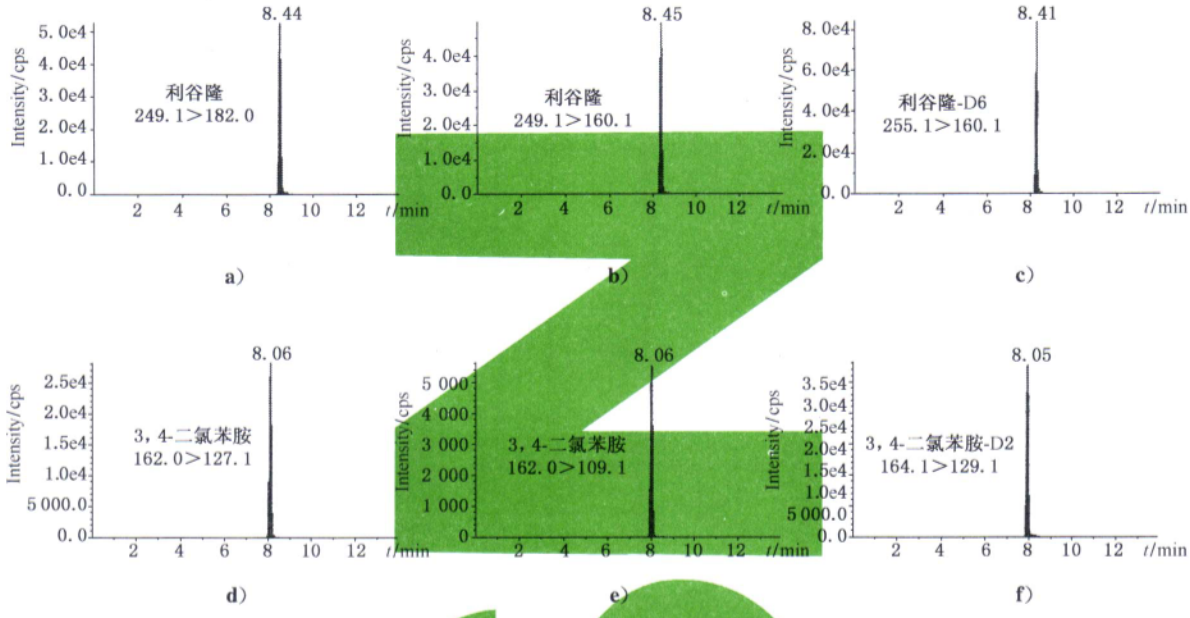

图 B.1 20 ng/mL 利谷隆和 3,4-二氯苯胺标准溶液的多反应监测(MRM)色谱图

附 录 C
（资料性附录）
肉及肉制品中利谷隆和3,4-二氯苯胺回收率范围

表 C.1 肉及肉制品中利谷隆和3,4-二氯苯胺添加回收率范围（$n=10$）

基质	化合物	添加水平 μg/kg	回收率 %	基质	化合物	添加水平 μg/kg	回收率 %
猪肉	利谷隆	10.0	77.0～102.0	猪肾	利谷隆	10.0	85.0～103.0
		50.0	86.4～102.0			100.0	88.3～110.3
		500.0	86.9～111.2			1 000.0	84.4～101.2
	3,4-二氯苯胺	10.0	84.0～106.0		3,4-二氯苯胺	10.0	71.0～107.0
		50.0	86.7～109.6			100.0	79.2～105.6
		500.0	81.5～110.2			1 000.0	87.1～111.4
猪肝	利谷隆	10.0	76.0～114.0	猪肠衣	利谷隆	10.0	88.0～105.0
		100.0	76.2～108.7			50.0	86.3～109.8
		1 000.0	92.3～108.8			500.0	82.6～109.7
	3,4-二氯苯胺	10.0	80.0～111.0		3,4-二氯苯胺	10.0	78.8～106.0
		100.0	82.6～101.5			50.0	88.9～106.8
		1 000.0	90.8～110.8			500.0	83.0～112.2
午餐肉	利谷隆	10.0	84.6～104.0	香肠	利谷隆	10.0	87.0～99.5
		50.0	86.1～113.8			50.0	86.2～109.3
		500.0	88.3～105.0			500.0	88.0～114.3
	3,4-二氯苯胺	10.0	78.0～112.0		3,4-二氯苯胺	10.0	84.6～107.0
		50.0	86.2～105.3			50.0	85.6～112.2
		500.0	88.8～111.4			500.0	90.7～105.0

Foreword

This standard was drafted under the rules derived from GB/T 1.1—2009.

The standard is for replace SN 0639—1997 "Method for determination of linuron residues in meat and meat products for export".

The main improvement from SN 0639—1997:

—The name of the standard is changed, and the metabolite residue is also determined;

—The scope of matrix is enlarged;

—The sampling processure is ommited;

—The method is changed to LC-MS/MS. This method makes the limit of quantification lower.

It is noted that some contents in this file may involve in patent authority. Agency regarding distribution and publication of this file does not be responsible for the identification of these patents.

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

The standard was drafted by Shanghai Entry-Exit Inspection and Quarantine Bureau of the People's Republic of China.

The standard was mainly drafted by Yi Xionghai, Qu Li, Cao Chen, Guo Dehua, Deng Xiaojun, Zhu Jian, Sheng Yonggang, Yang Huiqin, Bao Ming.

The original standard was SN 0639—1997 and this amendment for the first time.

Determination of linuron and its metabolite residues in meat and meat products for export—HPLC-MS/MS method

1 Scope

The standard specifies the determination of linuron and its metabolite residues in meat and meat products for export by HPLC-MS/MS.

The standard is applicable to the determination and confirmation of linuron and its metabolite residues in pork, liver, kidney, casings, canned steamed pork and sausage for export.,

2 Normative reference

The following documents for the application of standard is essential in this text. For dated references, only dated edition is suitable for this standard. For undated references, the latest edition (including all the amendments) is suitable for this standard.

GB/T 6682 Water for analytical laboratory use—Specification and test methods

3 Principle

The residues of linuron and its metabolite residues are axtracted by using combination solution of bacetone and acetonitrile. The extraction is defatted under −18 ℃ and cleaned up by Florisil solid phase extract column. Linuron and its metabolite residues are determined by HPLC-MS/MS, and quantified by internal standard method.

4 Reagents and materials

Unless otherwise specified, the entire reagent used should be analytical grade, water is deionizer water.

4.1 Acetonitrile: HPLC grade.

4.2 Acetone: HPLC grade.

4.3 Hexane: HPLC grade.

4.4 Diethylether: HPLC grade.

4.5 Formic acid: Purity≥99%.

4.6 Ammonlum acetate.

4.7 Acetone + acetonitrile (5 + 95, *V*1 + *V*2): Mix 50 mL of acetone with 950 mL of acetonitrile.

4.8 Diethylether + hexane(1 + 9, *V*1 + *V*2): Mix 100 mL of diethylether with 900 mL of hexane.

4.9 Acetone + hexane(1 + 9, *V*1 + *V*2): Mix 100 mL of acetone with 900 mL of hexane.

4.10 Acetonitrile + water(1 + 1, *V*1 + *V*2): Mix 100 mL of acetonitrile with 100 mL of water.

4.11 Anhydrous sodium sulfate: Dried at 650 ℃ for 4 h and keep in a tightly closed container.

4.12 5 mmol/L ammonium acetate + 0.1% formic acid: Weigh accurately 385.4 mg of ammonium acetate and add 1 mL of formic acid, add water to final volume of 1 L.

4.13 Standards: Linuron (CAS No: 330-55-2), Linuron-D6, 3, 4-Dichloroaniline (CAS No: 95-76-1), 3,4-Dichloroaniline-2,6-D2, purity of all chemicals was more than 98%.

4.14 Standard stock solution: Weigh accurately 0.01 g (accurate to 0.000 1 g) standard of linuron, Linuron-D6, 3, 4-Dichloroaniline, 3, 4-Dichloroaniline-2, 6-D2 and dissovle in acetonitrile to final concentration of 1 mg/mL respectively. All standards were refrigerated at −18 ℃, assign a shelflife of 12 months.

4.15 Stock standard solution of intermediate standards: Dilute stock standard solution to final concentration of 1 μg/mL in acetonitrile, All standards were refrigerated at −18 ℃, assign a shelflife of 6 months.

4.16 Calibration curve working solutions: Prepare the calibration curve solution at proper concentration by blank matrix. These working standard solutions are store refrigerated at 4 ℃, diluted directly before use.

4.17 Membrane filter: 0.45 μm, organic phase.

4.18 Florisil column: 6 mL, 1 g or equalvent. Condition the Florisil column with 5 mL hexane before use.

5 Apparatus and equipment

5.1 Liquid chromatography-mass/mass spectrometry, equipped with electrospray ion source and triquadruple mass spectrometer.

5.2 Electronic balance: Accurate to 0.000 1 g, 0.01 g.

5.3 Vortex mixer.

5.4 Centrifuge: 10 000 r/min.

5.5 Solid phase extraction set.

5.6 Nitrogen evaporator.

5.7 −20 ℃ refrigerator.

5.8 Plastic centrifuge tube: 50 mL.

5.9 Glass tube: 25 mL with graduation.

6 Preparation and storage

6.1 Preparation of test sample

The combined primary sample is reduced to 500 g which is totally minced and placed into a clean vessel as a test sample, which is sealed and labeled.

6.2 Storage of test sample

The test sample should be stored at temperature of below −18 ℃. Casing sample is desalted before using.

7 Procedure

7.1 Extraction

Weigh 2 g (accurate to 0.01 g) sample in 50 mL plastic centrifuge tubes, add 100 μL Linuron-D6 and 3,4-Dichloroaniline-2, 6-D inernal standard solution respectively (4.15), and add 2 g anhydrous sodium sulfate, mix well, then add 15 mL acetone + acetonitrile (5 + 95, $V1 + V2$) (4.7), homogenize

for 2 min, centrifuge at 3 000 r/min for 3 min, transfer the supematant to a new plastic centrifuge tube, and add another 10 mL acetone + acetonitrile (5 + 95, *V*1 + *V*2) to repeat the step above. Finally, combine the supernatant.

7.2 Cleaning-up

Put the extracts in −18 ℃ refrigerator for 1 h, then filter into a glass graduated tube quickly, and add acetone to make the final volume at 25 mL then accurately transfer 5 mL solution to a clean tube. The solution is concentrated to nearly dryness by gentle nitrogen under room temperature. Then add 2 mL of hexane to each tube, mix well.

Load the sample solution to the Florisil columns, every tube is washed with 2 mL of diethylether + hexane (1 + 9, *V*1 + *V*2) (4.8) and load to the Florisil columns. Every column is washed by 4 mL diethylether + hexane (1 + 9, *V*1 + *V*2). Compounds of interest are eluted three times by acetone + hexaue (1 + 9, *V*1 + *V*2) (4.9) and finally volume is 7.5 mL. Generally, the flow rate should not exceed 30 d/min. The elute is collected into a glass tube and is concentrated to nearly dryness by gentle nitrogen under 40 ℃ and add 1 mL of acetonitrile + water (1 + 1, *V*1 + *V*2) (4.10) to resolve the residence and filter by 0.45 μm film before HPLC-MS/MS analysis.

7.3 Determination

7.3.1 HPLC operating conditions

The HPLC operating conditions are as follows:

a) LC column: C_{18} column, 50 mm × 4.6 mm (i.d), 1.8 μm (of other equivalent ones);

b) Mobile phase: A: 5 mmol/L ammonium acetate + 0.1% formic acid; B: acetonitrile, the elution gradient is listed in Table 1;

Table 1—Mobile phase and elution gradient

Time/min	Acetonitrile/%	5 mmol/L Ammonium acetate + 0.1% Formic acid/%
0.0	2	98
2.0	2	98
5.5	99	1
11.1	99	1
11.2	2	98
18.0	2	98

c) Flow rate:0.3 mL/min;

d) Injector volume:20 μL;

e) Column temperature:Room temperature.

7.3.2 Mass spectral acquisition

The Mass spectral acquistion conditions are as follows:

a) Ion source:ESI;

b) Monitoring model:Multiple reaction monitoring (MRM);

c) Nebulizer gas (GS1),curtain gas (CUR),auxiliary heater gas (GS2) and collision gas are high purity nitrogen or equivalent,optimize the flow rate of each gas and ion source temperature to reach the equirment of the sensitivity of mass spectrometry.Detailed parameters are shown as annex A;

d) Collision energy (CE),deculstering potential (DP),collision cell exit potential (CXP),collision cell entrance potential (EP) and electros pray capillary voltage (IS) should be optimized to the best sensitivity.Related parameters and qualifier and quantifier MRM are listed as Table A.1 in annex A.

7.3.3 Quantitation of HPLC-MS/MS

According to established HPLC-MS/MS operating condition,determine the sample solution and the standard working curve simultaneously.The standard working curve should contained 5 level of concentration including the zero point.If the determined sample is over the scope of standard working curve,the concentration of determined sample should be diluted to a proper concentration by mobile phase.Under the above HPLC-MS/MS operating condition,the retention time of linuron and 3,4-dichloroaniline are 8.5 min,8.0 min respectively,the MRM chromatograms of the standard are listed as annex B figure B.1.

7.3.4 Confirmation of HPLC-MS/MS

Determinate under the established HPLC-MS/MS conditions,and calculated the intensity ration of two selected ion pairs of the sample solution and the standard working solution.If the retention times of sample chromatogram peaks are consistent with that of working solution and their windage is less than 5%,and the relative abundance ratio tolerance is the same as listed (table 2),it is safe to conclude that this compound exit in the sample.

Table 2—Maximum permitted tolerances for relative ion intensities while conformation %

Relative ion intensities	>50	20～50	10～20	≤10
Permitted relative tolerances	±20	±25	±30	±50

7.4 Blank test

The operation of the blank test is the same as describe in the method of determination, but without addition the sample.

8 Calculation and expression of result

Calculate the content of benzodiazepine residues in the test sample by HPLC-MS/MS data processor or according to the formula (1). The blank value should be, subtracted from the above result of calculation.

$$X_i = \frac{R_i \times c_i \times V}{R_s \times m} \times \frac{1\,000}{1\,000} \quad \cdots\cdots (1)$$

where

X_i —the residue content of linuron or 3,4-dichloroaniline, μg/kg;

R_i —the ration of peak areas between analytes and internal standard in sample solution;

c_i —the concentration of linuron or 3,4-dichloroaniline residue from standard working curve, ng/mL;

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

R_s —the ration of peak areas between analytes and intemal standards in calbiration curves;

m —mass of test sample of final sample solution, g.

9 Limit of quantification (LOQ) and recovery

9.1 Limit of quantification (LOQ)

The limit of quantitation is 10 μg/kg for linuron and 3,4-dichloroaniline.

9.2 Recovery

According to this method, add standard solutions in eight materials: pork, liver, kidney, casings, canned steamed pork and sausage, the recovery ranges of linuron and 3,4-dichloroaniline in eight base materials are listed as table C.1 in annex C.

Annex A
(informative)
Main mass parameters of API 4000[1)]

Main mass parameters:

a) Electros pray capillary voltage (IS): positive mode 5 500 V;

b) CAD: Medium;

c) GS1: 517 kPa (70 psi);

d) CUR: 172 kPa (30 psi);

e) GS2: 414 kPa (60 psi);

f) TEM: 550 ℃;

g) Precursor ions, Collision Energy (CE), Declustering Potential (DP), collision cell entrance potential (EP), collision cell exit potential (CXP), as Table A.1.

Table A.1—Transitions for confirmation and quantification of linuron and 3,4-dichloroaniline CE、DP、EP、CXP

Compound	Precursor ion(Q1)	Product ion(Q3)	CE (V)	DP/V	EP/V	CXP/V
linuron	249.1	160.1*	24.5	60.0	9.0	13.0
		182.0	22.0	60.0	9.0	11.0
linuron-D6	255.1	160.1*	23.0	64.0	12.0	9.0
3,4-dichloroaniline	162.0	109.1	44.0	67.0	8.0	21.0
		127.1*	28.0	67.0	8.0	25.0
3,4dichloroaniline-2,6-D2	164.1	129.1*	28.5	60.0	6.0	25.0
Annotation: the symbol"*"represents the quantitative transition.						

1) Non-commercial statement: the equipment and their types involved in the standard method are not related to commercial aims, and it is encouraged to use equipment of different corporation or different type.

Annex B
(informative)
MRM chromatogram of linuron and 3,4-dichloroaniline standards

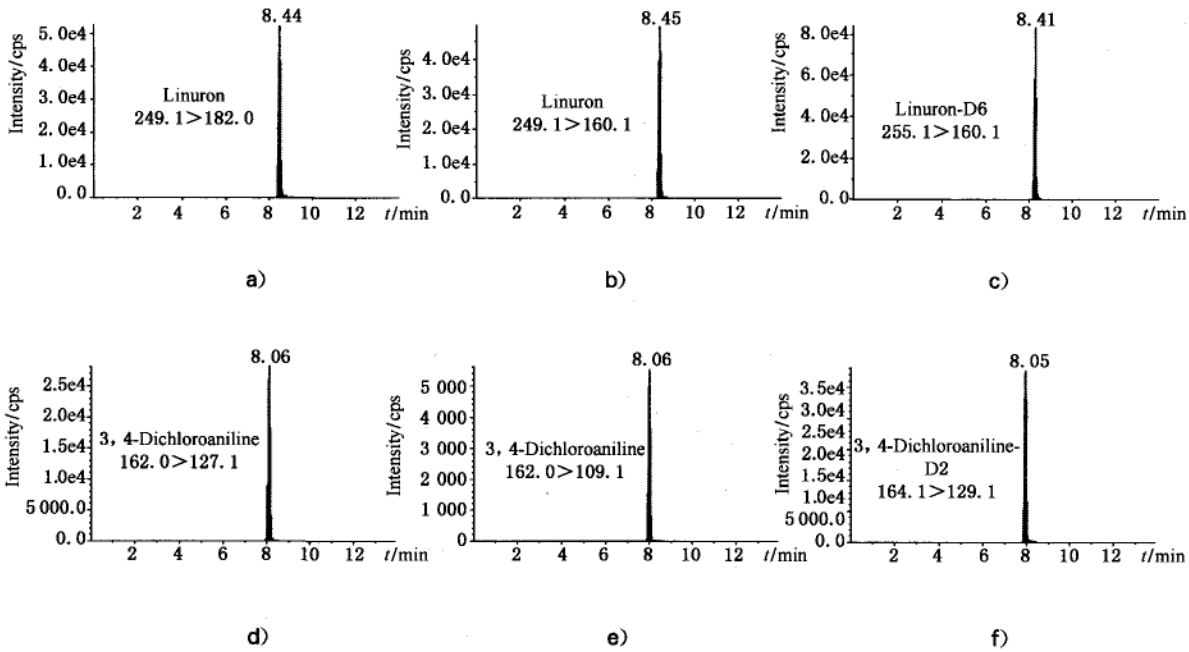

Figure B.1—MRM chromatogram of linuron (20 ng/mL) and 3,4-dichloroaniline (20 ng/mL) standards

Annex C
(informative)
Recovery ranges of linuron and 3,4-dichloroaniline in meat and meat products

Table C.1—Recovery ranges of linuron and 3,4-dichloroaniline in meat and meat products

Matrix	Residuum	Fortified level μg/kg	Recovery %	Matrix	Residuum	Fortified level μg/kg	Recovery %
pork	linuron	10.0	77.0～102.0	liver	linuron	10.0	85.0～103.0
		50.0	86.4～102.0			100.0	88.3～110.3
		500.0	86.9～111.2			1 000.0	84.4～101.2
	3,4-dichloroaniline	10.0	84.0～106.0		3,4-dichloroaniline	10.0	71.0～107.0
		50.0	86.7～109.6			100.0	79.2～105.6
		500.0	81.5～110.2			1 000.0	87.1～111.4
kidney	linuron	10.0	76.0～114.0	casings	linuron	10.0	88.0～105.0
		100.0	76.2～108.7			50.0	86.3～109.8
		1 000.0	92.3～108.8			500.0	82.6～109.7
	3,4-dichloroaniline	10.0	80.0～111.0		3,4-dichloroaniline	10.0	78.8～106.0
		100.0	82.6～101.5			50.0	88.9～106.8
		1 000.0	90.8～110.8			500.0	83.0～112.2
canned steamed pork	linuron	10.0	84.6～104.0	sausage	linuron	10.0	87.0～99.5
		50.0	86.1～113.8			50.0	86.2～109.3
		500.0	88.3～105.0			500.0	88.0～114.3
	3,4-dichloroaniline	10.0	78.0～112.0		3,4-dichloroaniline	10.0	84.6～107.0
		50.0	86.2～105.3			50.0	85.6～112.2
		500.0	88.8～111.4			500.0	90.7～105.0