



## Experience Sharing Seminar for Food Testing Laboratories (Online)

### Laboratory preparation for the implementation of The Harmful Substances in Food (Amendment) Regulation 2021 (Cap. 132AF) and Testing of mycotoxins

8 Dec 2022

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**Government Laboratory,**

The Government of the Hong Kong Special Administrative Region

Metrology Forensic Toxicology 環境衛生 藥物安全 Questioned Documents Public Safety  
Contact Evidence 法證毒理 商品說明 Science 罪案現場調查 食物安全 科學服務 CONTROLLED DRUGS  
Drug Safety 公眾安全 Trade Description Forensic DNA 環境保護 化學計量 受管制藥物 中藥分析 法證DNA 接觸證據 ENVIRONMENTAL HYGIENE  
Environmental Protection Chinese Medicine Analysis Crime Investigation 文件鑑辨



# 《2021年食物內有害物質(修訂)規例》

## The Harmful Substances in Food (Amendment) Regulation 2021 (Cap. 132AF)

### 真菌毒素檢測

### Testing of mycotoxins



# 食物內有害物質規例 (第132 AF章)

## The Harmful Substances in Food Regulation (Cap. 132AF)

有害物質  
Harmful  
substances

獸藥殘餘  
Veterinary drugs  
residues



# 食物內有害物質規例 (第132 AF章)

## The Harmful Substances in Food Regulation (Cap. 132AF)

### 有害物質 Harmful substances

黃曲霉毒素  
Aflatoxins

三聚氰胺  
Melamine

孔雀石綠  
Malachite Green

芥酸  
Erucic acid



# 2021年食物內有害物質(修訂)規例 (第132 AF章)

## The Harmful Substances in Food (Amendment) Regulation 2021 (Cap. 132AF)

實施日期 2023 年 6 月 1 日  
1 June 2023 come into operation

實施日期 2023 年 12 月 1 日  
1 December 2023 come into operation

### 有害物質 Harmful substances

法律公告編號 L.N. 86 of 2021

孔雀石綠  
Malachite Green

芥酸  
Erucic acid

黃曲霉毒素  
Aflatoxins

脫氧雪腐鏟刀菌烯醇  
Deoxynivalenol

三聚氰胺  
Melamine

棒曲霉毒素  
Patulin

苯並[a]芘  
Benzo[a]pyrene

3-氯-1,2-丙二醇  
3-MCPD

縮水甘油脂肪酸酯  
Glycidyl fatty acid esters

部分氫化油  
Partially hydrogenated oil  
(PHO)



# ~~2021~~年食物內有害物質(修訂)規例 (第132 AF章)

## The Harmful Substances in Food (Amendment) Regulation ~~2021~~ (Cap. 132AF)



### 政策措施 Policy Measures 2022

在完成檢視有關獸藥殘餘的食物安全標準後，於2023年就擬議的規管標準諮詢公眾。

Conduct public consultation on the proposed regulatory standards relating to veterinary drug residues in food in 2023, upon completion of the review on relevant food safety standards.

### 獸藥殘餘 Veterinary drugs residues





黃曲霉毒素  
Aflatoxins

# 現行規例.....

## Current Regulation.....

<u>物質</u> <u>Substance</u>	<u>物質之描述</u> <u>Description of substance</u>	<u>食物類別</u> <u>Description of food</u>	<u>最高濃度</u> <u>Maximum concentration</u>
黃曲霉毒素 Aflatoxins	雙呋喃氧雜萜鄰酮屬的化合物，並包括黃曲霉毒素 B1、B2、G1、G2、M1、M2、P1 及黃曲霉毒素醇 Group of bisfuranocoumarin compounds and includes aflatoxin B1, B2, G1, G2, M1, M2, P1 and aflatoxicol	花生或花生產品 Peanuts or peanut Products	20 µg/kg
		任何食物( 花生或花生產品除外) Any food other than peanut or its products	15 µg/kg



黃曲霉毒素  
Aflatoxins

# 修訂規例..... Amendment Regulation.....

物質 Substance	物質之描述 Description of substance	食物類別 Description of food	最高濃度 Maximum concentration
總黃曲霉毒素 Aflatoxins, total	黃曲霉毒素 B1、B2、G1、G2 之和 Sum of aflatoxin B1, B2, G1 and G2	非即食的杏仁、巴西堅果、榛子、花生及開心果 Non-ready-to-eat almonds, Brazil nuts, hazelnuts, peanuts and pistachios	15 µg/kg
		非即食的花生產品及杏仁、巴西堅果、榛子及開心果產品 Non-ready-to-eat peanut products and products of almonds, Brazil nuts, hazelnuts and pistachios	
		香料 Spices	10 µg/kg
		任何其他食物 Any other food	





黃曲霉毒素  
Aflatoxins

# 修訂規例.....

## Amendment Regulation.....

物質  
Substance

食物類別  
Description of food

最高濃度  
Maximum concentration

黃曲霉毒素 B1  
Aflatoxin B1

嬰兒配方產品及較大嬰兒及幼兒配方產品 (以奶類蛋白質製造的配方產品除外)  
Infant formula and follow-up formula **other than formula manufactured from milk protein**

0.1 µg/kg  
(Note 1)

任何其他擬主要供不足 36 個月大的人食用的食物 (以奶類蛋白質製造的嬰兒配方產品及較大嬰兒及幼兒配方產品除外)  
Any other food intended to be consumed principally by persons under the age of 36 months **other than infant formula and follow-up formula manufactured from milk proteins**

0.1 µg/kg  
(Note 2)

黃曲霉毒素 M1  
Aflatoxin M1

擬主要供不足12 個月大的人食用的嬰兒配方產品及較大嬰兒及幼兒配方產品  
Infant formula and follow-up formula intended to be consumed principally by persons under the age of 12 months

0.025 µg/kg  
(Note 1)

任何其他奶類或奶粉  
Any other milk or dried milk

0.5 µg/kg  
(Note 1)

Note 1: 最高濃度適用於處於或已調配至可即時食用狀態的食物。The maximum concentration applies to the food that is, or is reconstituted to be, ready for consumption.

Note 2: 最高濃度適用於食物的乾物質 The maximum concentration applies to the dry matter of the food.



三聚氰胺  
Melamine

# 現行規例..... Current Regulation.....

食物類別  
Description of food

最高濃度  
Maximum concentration

奶類  
Milk

擬主要供涵蓋36個月以下幼兒的某年齡組別的人食用的任何食物

Any food intended to be consumed principally by persons of an age group into which children under the age of 36 months fall

擬主要供懷孕或授乳的女性食用的任何食物

Any food intended to be consumed principally by pregnant or lactating women

任何其他食物  
Any other food

1 mg/kg

2.5 mg/kg



三聚氰胺  
Melamine

# 修訂規例.....

## Amendment Regulation.....

食物類別

Description of food

最高濃度

Maximum concentration

擬主要供不足 12 個月大的人食用的液態嬰兒配方產品及液態 較大嬰兒及幼兒 配方產品

Liquid infant formula and liquid follow-up formula intended to be consumed principally by persons under the age of 12 months

} 0.15 mg/kg

奶類 (擬主要供不足12個月大的人食用的液態嬰兒配方產品及液態較大嬰兒及幼兒配方產品除外 )

Milk other than liquid infant formula and liquid follow-up formula intended to be consumed principally by persons under the age of 12 months

擬主要供涵蓋36 個月以下幼兒的某年齡組別的人食用的任何食物

Any food intended to be consumed principally by persons of an age group into which children under the age of 36 months fall

} 1 mg/kg

擬主要供懷孕或授乳的女性食用的任何食物

Any food intended to be consumed principally by pregnant or lactating women

任何其他食物

Any other food

} 2.5 mg/kg

芥酸  
Erucic acid

# 現行規例..... Current Regulation.....

<u>物質</u> <u>Substance</u>	<u>物質之描述</u> <u>Description of substance</u>	<u>食物類別</u> <u>Description of food</u>	<u>最高濃度</u> <u>Maximum concentration</u>
芥酸 Erucic acid	脂肪酸順(式) 13-二十二(碳)烯酸 The fatty acid cis-docos-13-enoic acid	任何油或脂肪或兩者的混合物 Any oil or fat or any mixture thereof  加有油或脂肪或加有兩者的混合物的任何食物 Any food to which oil or fat or a mixture thereof has been added	以重量計其所含脂肪酸的 5% 5 % by weight of their fatty acid content  以重量計食物內全部油及脂肪所含脂肪酸的 5% 5% by weight of their fatty acid content of all the oils and fats in the food.



芥酸  
Erucic acid

# 修訂規例..... Amendment Regulation.....

<u>物質</u> <u>Substance</u>	<u>物質之描述</u> <u>Description of substance</u>	<u>食物類別</u> <u>Description of food</u>	<u>最高濃度</u> <u>Maximum concentration</u>
		低芥酸菜籽油 Low erucic acid rapeseed oil	以重量計其所含脂肪酸的 2% 2 % by weight of their fatty acid content
芥酸 Erucic acid	脂肪酸順(式) 13-二十二(碳)烯酸 The fatty acid cis-docos-13-enoic acid	任何其他油或脂肪或任何油及脂肪的混合物 Any other oil or fat or any mixture of oil and fat  加有油或脂肪或兩者的混合物的食物 Any food to which oil or fat or a mixture thereof has been added	以重量計其所含脂肪酸的 5% 5 % by weight of their fatty acid content  以重量計食物內全部油及脂肪所含脂肪酸的 5% 5% by weight of their fatty acid content of all the oils and fats in the food.





脫氧雪腐鏟刀菌烯醇  
Deoxynivalenol

棒曲霉素  
Patulin

# 修訂規例.....

## Amendment Regulation.....

物質  
**Substance**

食物類別  
**Description of food**

最高濃度  
**Maximum concentration**

脫氧雪腐鏟刀菌烯醇  
Deoxynivalenol

擬主要供不足 36 個月大的人食用的穀基類食物  
Cereal-based foods intended to be consumed principally  
by persons under the age of 36 months

200 µg/kg  
(Note 2)

棒曲霉素  
Patulin

蘋果汁及加有蘋果汁的其他飲品  
Apple juice and other beverages to which apple juice has  
been added

50 µg/kg  
(Note 1)

Note 1: 最高濃度適用於處於或已調配至可即時食用狀態的食物。The maximum concentration applies to the food that is, or is reconstituted to be, ready for consumption.

Note 2: 最高濃度適用於食物的乾物質 The maximum concentration applies to the dry matter of the food.





3-氯-1,2-丙二醇  
3-MCPD

苯並[a]芘  
Benzo[a]pyrene

# 修訂規例..... Amendment Regulation.....

<u>物質</u> Substance	<u>食物類別</u> Description of food	<u>最高濃度</u> Maximum concentration
3-氯-1,2-丙二醇 3-MCPD	含有酸水解植物蛋白的固態調味品 Solid condiments containing acid hydrolysed vegetable proteins	1 mg/kg
	任何其他含有酸水解植物蛋白的調味品 Any other condiments containing acid hydrolysed vegetable proteins	0.4 mg/kg
苯並[a]芘 Benzo[a]pyrene	油或脂肪或兩者的混合物 Oil or fat or any mixture of oil and fat	5 µg/kg
	擬主要供不足12個月大的人食用的嬰兒配方產品及較大嬰兒及幼兒配方產品 Infant formula and follow-up formula intended to be consumed principally by persons under the age of 12 months	1 µg/kg



縮水甘油脂肪酸酯  
Glycidyl fatty acid esters

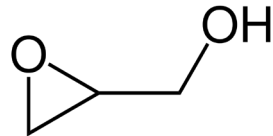
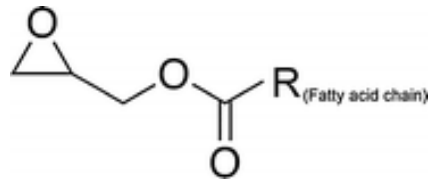
# 修訂規例.....

## Amendment Regulation.....

物質 <u>Substance</u>	物質之描述 <u>Description of substance</u>	食物類別 <u>Description of food</u>	最高濃度 <u>Maximum concentration</u>
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縮水甘油脂肪酸酯  
Glycidyl fatty acid  
esters

縮水甘油脂肪酸酯，  
以環氧丙醇表示  
Glycidyl fatty acid  
esters expressed  
as glycidol



擬主要供不足12 個月大的人食用的粉狀  
嬰兒配方產品及粉狀較大嬰兒及幼兒配方  
產品

Powdered infant formula and powdered  
follow-up formula intended to be  
consumed principally by persons under  
the age of 12 months

50 µg/kg

擬主要供不足12 個月大的人食用的液態嬰兒  
配方產品及液態較大嬰兒及幼兒配方產品

Liquid infant formula and liquid follow-up  
formula intended to be consumed  
principally by persons under the age of 12  
months

6 µg/kg



部分氫化油  
Partially hydrogenated oil  
(PHO)

# 修訂規例.....

## Amendment Regulation.....

部分氫化油指有經過氫化的過程但並無因為該過程而完全飽和的油或脂肪

Partially hydrogenated oil means any oil or fat that has undergone the process of hydrogenation but is not fully saturated as a result of that process.

# 違禁物質

## Prohibited substance



# 《2021年食物內有害物質(修訂)規例》

## The Harmful Substances in Food (Amendment) Regulation 2021 (Cap. 132AF)

### 真菌毒素檢測

### Testing of mycotoxins



# 真菌毒素檢測

## Testing of mycotoxins

黃曲霉毒素  
Aflatoxins

脫氧雪腐鐮刀菌烯醇  
Deoxynivalenol

棒曲霉毒素  
Patulin



中華人民共和國香港特別行政區政府  
The Government of the Hong Kong Special Administrative Region  
of the People's Republic of China

ENGLISH | 繁體版 | 简体版 | 郵件





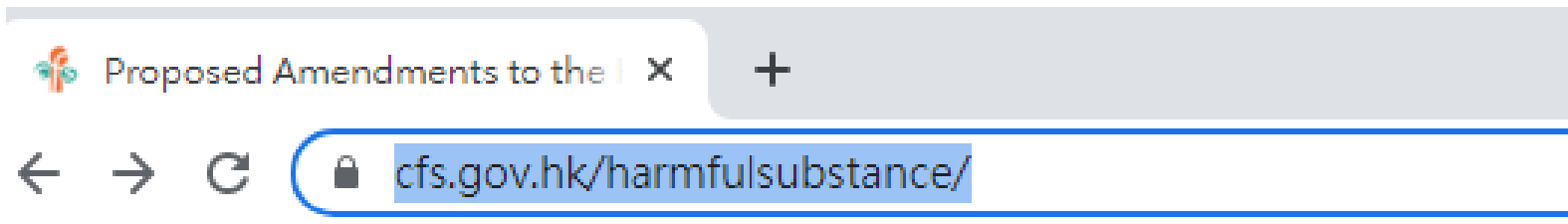
# 真菌毒素檢測

## Testing of mycotoxins

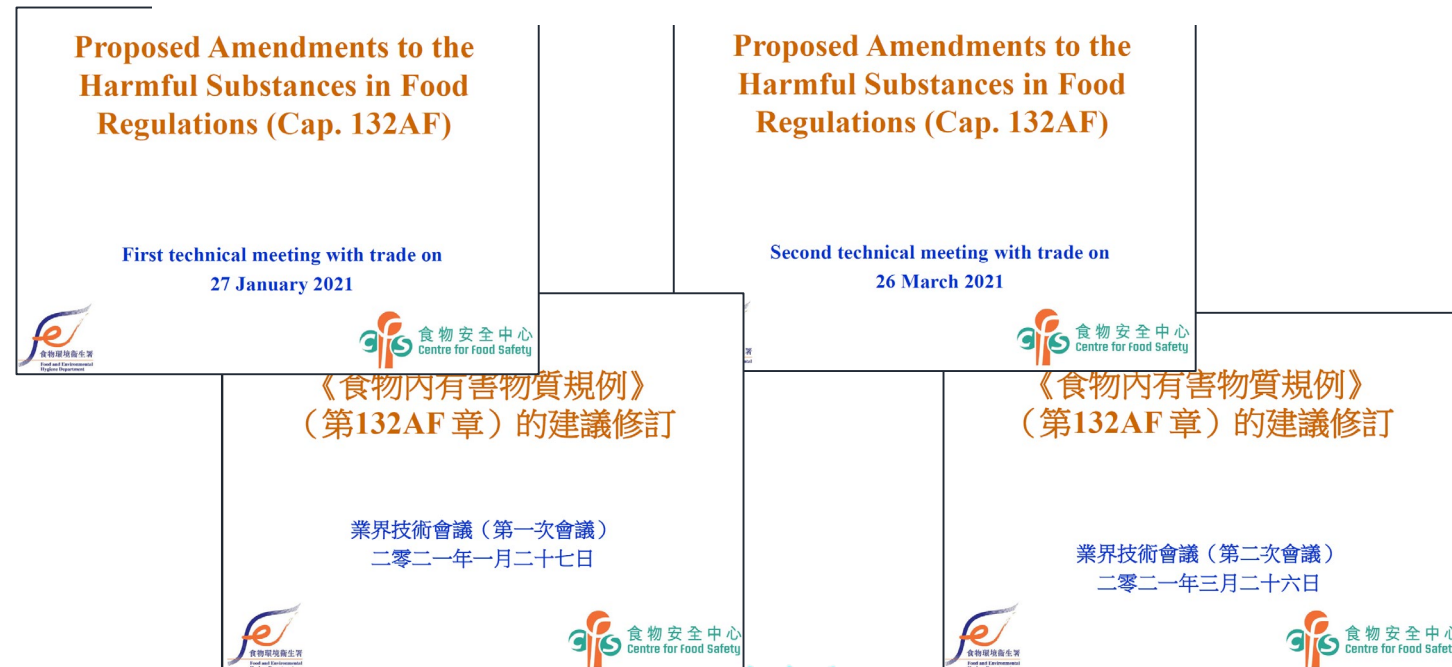
黃曲霉毒素  
Aflatoxins

脫氧雪腐镰刀菌烯醇  
Deoxynivalenol

棒曲霉毒素  
Patulin



中華人民共和國香港特別行政區  
The Government of the Hong Kong Special Administrative Region  
of the People's Republic of China





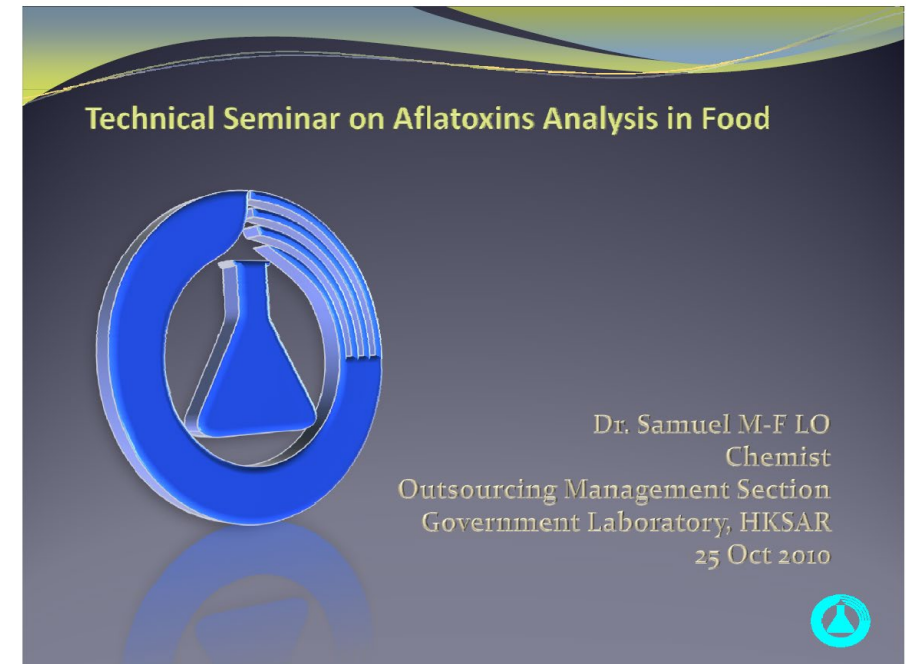
# 真菌毒素檢測

## Testing of mycotoxins

黃曲霉毒素  
Aflatoxins

脫氧雪腐鐮刀菌烯醇  
Deoxynivalenol

棒曲霉毒素  
Patulin



# 真菌毒素檢測

## Testing of mycotoxins

黃曲霉毒素  
Aflatoxins

脫氧雪腐鐮刀菌烯醇  
Deoxynivalenol

棒曲霉毒素  
Patulin

govtlab.gov.hk/tc/our\_work/development.html

香港特別行政區政府  
政府化驗所

輸入查詢字串

家 最新消息 關於我們 我們的工作 一般資訊 聯絡我們

為檢測業界提供技術交流會

簡介	日期
<a href="#">檢測水產食品中的硝基呋喃類代謝物</a>	2021年5月31日
<a href="#">食品過敏原分析方法概覽</a>	2020年6月29日
<a href="#">食品中霉菌毒素的檢測</a>	2020年3月13日
<a href="#">有關《2018年食物攪雜（金屬雜質含量）（修訂）規例》的開發工作</a>	2019年2月5日
<a href="#">有關《2018年食物攪雜（金屬雜質含量）（修訂）規例》的微量元素</a>	
食品中污染物的化學檢測技術研討會(與香港認可處合辦): (i) <a href="#">食品中二噁英及二噁英樣多氯聯苯的檢測和</a> (ii) <a href="#">食品中甲基汞和無機砷的檢測</a>	

測試方法

以下為政府化驗所在技術交流會上曾和業界分享與外判服務相關的測試方法摘要：

黃曲霉毒

Determination of Mycotoxins in Food

Government Laboratory

[https://www.govtlab.gov.hk/tc/our\\_work/development.html](https://www.govtlab.gov.hk/tc/our_work/development.html)



# 真菌毒素檢測

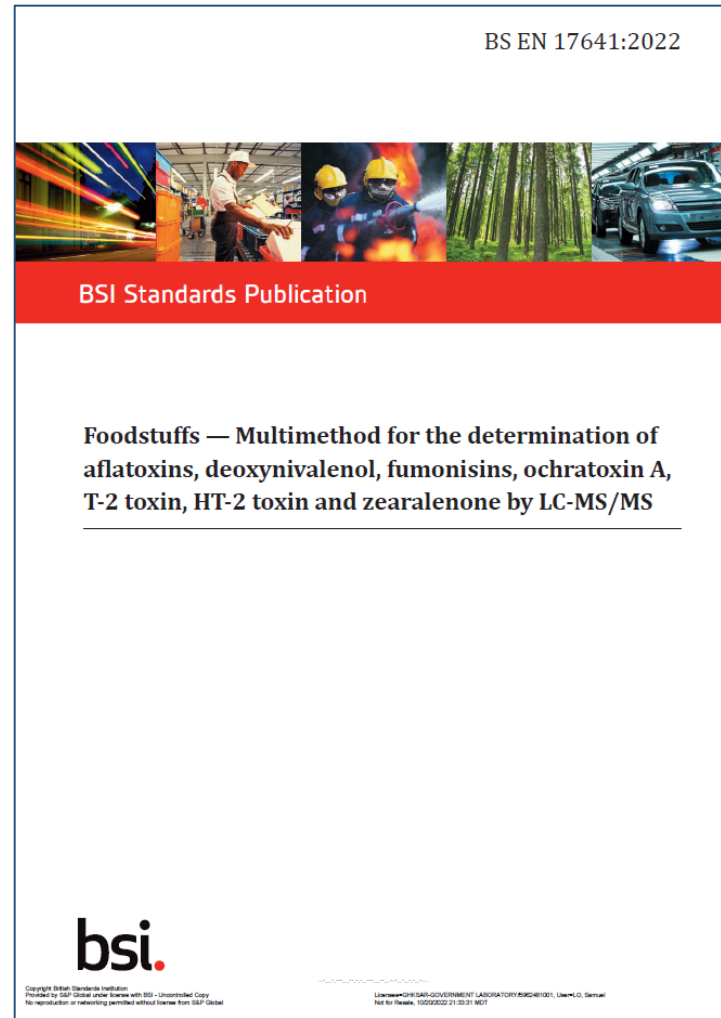
## Testing of mycotoxins

黃曲霉毒素  
Aflatoxins

脫氧雪腐鐮刀菌烯醇  
Deoxynivalenol

*Recently release.....*

EN 17641:2022



# 真菌毒素檢測

## Testing of mycotoxins

黃曲霉毒素  
Aflatoxins

本簡報所提供的資料是為方便出席者而設，並不構成政府化驗所對所指產品的認可。

脫氧雪腐鏟刀菌烯醇  
Deoxynivalenol

This information given in this presentation is for the convenience of attendees and does not constitute an endorsement by Government Laboratory of the product named.

棒曲霉毒素  
Patulin



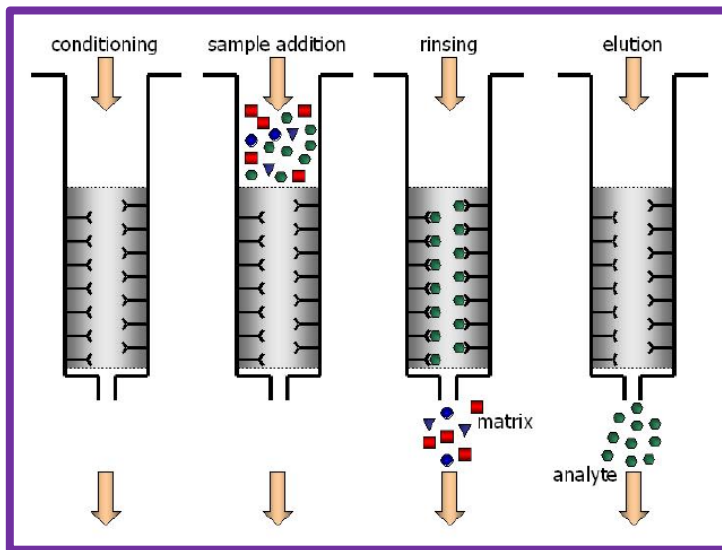
# 真菌毒素檢測

## Testing of mycotoxins

黃曲霉毒素  
Aflatoxins

脫氧雪腐鐮刀菌烯  
Deoxynivalenol

棒曲霉毒素  
Patulin



免疫親和柱  
Immunoaffinity column

提取 Extraction

脫脂 De-fat

固相萃取淨化 SPE Clean-up

黃曲霉毒素  
Aflatoxins  
*AFLAPREP®*

黃曲霉毒素 M1  
Aflatoxin M<sub>1</sub>  
*Afla M1™*

脫氧雪腐鐮刀菌烯  
Deoxynivalenol  
*Bond Elut  
Mycotoxin*

棒曲霉毒素  
Patulin  
*OASIS® HLB*

高效液相色譜或液相色譜串聯質譜分析  
HPLC or LC-MS/MS analysis





# 真菌毒素檢測

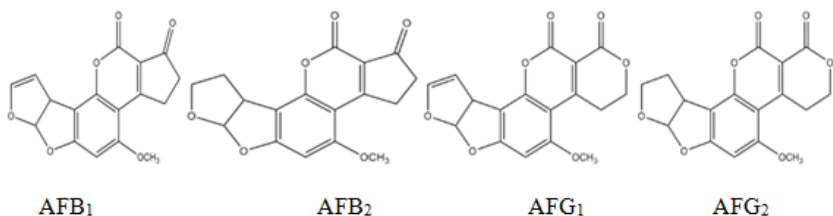
## Testing of mycotoxins

### 黃曲霉毒素 Aflatoxins

Reference to  
AOAC 991.31

25 g sample + 5 g NaCl + 125 mL 70% Methanol  
Blend and filter  
Pipette 15 mL filtrate + 30 mL water.  
(Adjust the MeOH < 10%).

25 g 樣品 + 5 g NaCl + 125 mL 70% 甲醇  
混合和過濾  
抽取15 mL 濾液 + 30 mL 水  
(調整 MeOH < 10% )

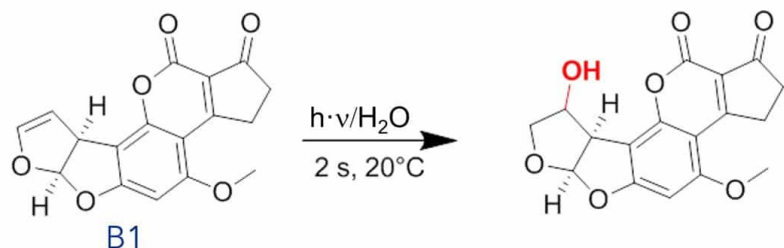


#### Immunoaffinity SPE Extraction

- Load 15 mL sample solution
- Wash with 10 mL water x 2
- Eluate with 1 mL MeOH
- Transfer all eluate to 2-mL V. flask and make up with water

#### 免疫親和固相萃取

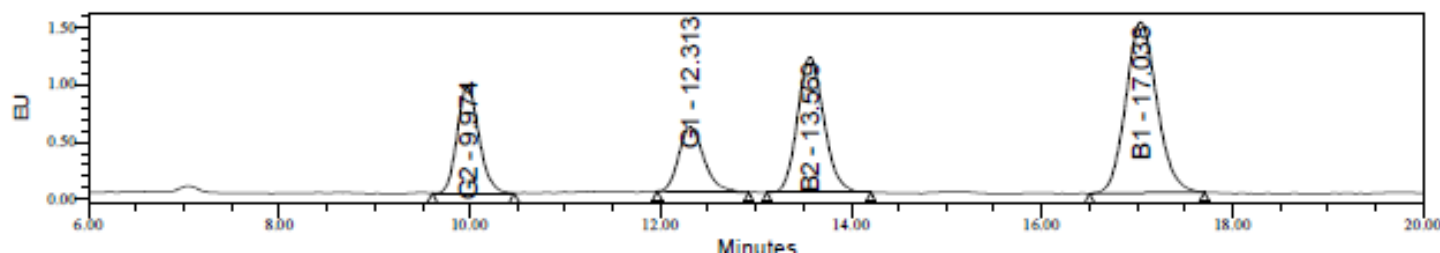
- 加載 15 mL 樣品溶液
- 用 10 mL 水洗滌兩次
- 用 1 mL 甲醇洗脫
- 將所有洗脫液轉移到 2-mL 容量瓶中並用水補足



高效液相色譜 (HPLC) 光化學柱後衍生法作測定

HPLC analysis with Post-column Photochemical Derivatization

應對每批次的親和柱進行質量驗證  
*Performance should be checked for each batch of columns*



報告限 Reporting limit = 1 µg/kg (total)

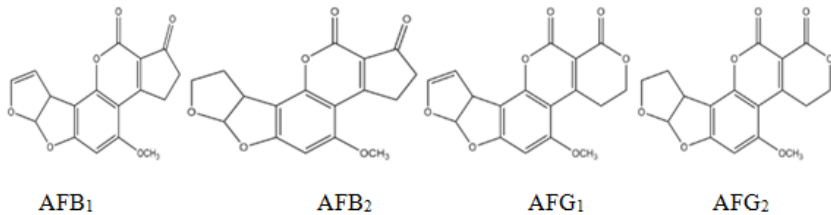


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## Testing of mycotoxins

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### 黃曲霉毒素 Aflatoxins



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混合和過濾  
抽取15 mL 濾液 + 30 mL 水  
(調整 MeOH < 10% )

#### Immunoaffinity SPE Extraction

- Load 15 mL sample solution
- Wash with 10 mL water x 2
- Eluate with 1 mL MeOH
- Transfer all eluate to 2-mL V. flask and make up with water

#### 免疫親和固相萃取

- 加載 15 mL 樣品溶液
- 用 10 mL 水洗滌兩次
- 用 1 mL 甲醇洗脫
- 將所有洗脫液轉移到 2-mL 容量瓶中並用水補足

加入同位素內標  
Spike isotope labelled internal standard

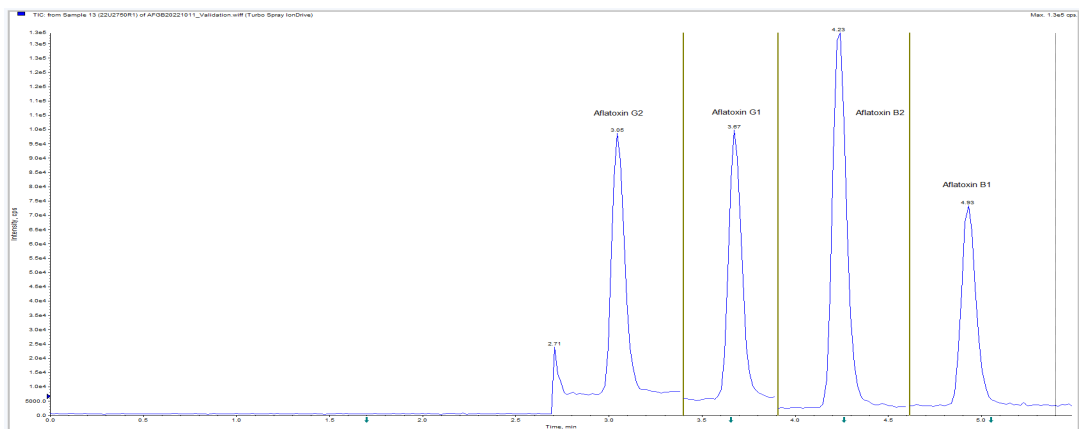
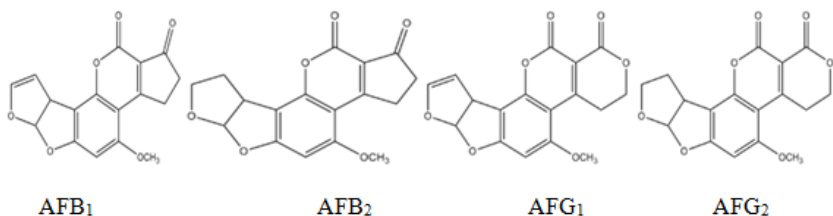
應對每批次的親和柱進行質量驗證  
*Performance should be checked for each batch of columns*

液相色譜串聯質譜分析  
LC-MS/MS analysis



# 真菌毒素檢測 Testing of mycotoxins

## 黃曲霉毒素 Aflatoxins



添加了0.01 ppb 嬰兒配方奶粉的液相串聯質譜色譜  
LC-MS/MS Chromatograms spike at 0.01 ppb (infant formula)

5 g 樣品  
5 g sample

+

加入同位素內標  
Spike isotope labelled internal standard

嬰兒配方奶粉  
Infant Formula

- + 20 mL 70% MeOH
- Ultrasonic 20 mins
- Centrifuge 10000 rpm 10 mins, filter
- Dry the solvent
- Reconstitute with 0.5 mL MeOH and 3.5 mL H<sub>2</sub>O
- Load all solution to SPE cleanup

- + 20 mL 70% 甲醇
- 超聲波振蕩20分鐘
- 離心 10000 rpm 10 分鐘,過濾
- 吹乾溶劑
- 用 0.5 mL 甲醇和 3.5 mL 水復溶
- 加載所有樣品溶液作固相萃取

Immunoaffinity SPE  
Extraction

免疫親和固相萃取

液相色譜串聯質譜分析  
LC-MS/MS analysis

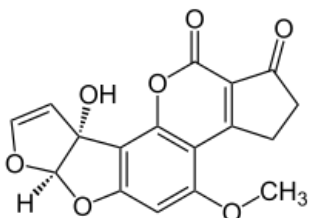
定量限 LOQ = 0.01 µg/kg



# 真菌毒素檢測 Testing of mycotoxins

## 黃曲霉毒素 Aflatoxins

### Aflatoxin M1



奶 Fluid milk

50 g 樣品  
50 g sample



- Centrifuge at 10000 rpm for 10 minutes at about 4°C to defat
- Remove the fat (top) layer
- Proceed to clean-up by IAC with all sample solution.

奶粉, 忌廉, 乳酪

Milk powder, cream and yogurt

25 g 樣品  
25 g sample

Add 80 mL hot water  
and mix

加入80 mL 熱水  
並攪均

- 在4°C 中離心 10000 rpm 10 分鐘 以脫脂
- 去除脂肪 (頂層) 層
- 用所有樣品溶液進行IAC萃取

芝士 Cheese

25 g 樣品  
25 g sample



- Add 100 mL dichloromethane and blend for 3 minutes
- Filter, and remove the solvent by rotary evaporator to just dryness
- Reconstituting the residue by dissolving in 5 mL of methanol, 10 mL of heptane and then 30 mL of water by order, and vortex
- Centrifuge at 10000 rpm for 5 minutes
- Collect the lower aqueous layer, Use separating funnel if necessary.
- Further extract the remaining organic layer by 10 mL water,
- Collect the lower aqueous layer.
- 加入 100 mL 二氯甲烷並用均質器混勻 3 分鐘
- 過濾, 並用旋轉蒸發溶液
- 依次用 5 mL 甲醇、10 mL 庚烷和 30 mL 水溶解殘留物, 然後渦旋均勻
- 離心10000 rpm 5 分鐘
- 將全部液體移至分液漏斗, 收集下層水層。
- 進一步用 10 mL 水萃取剩餘的有機層
- 收集所有下層水。

Immunoaffinity SPE Extraction

免疫親和固相萃取

加入同位素內標

Spike isotope labelled internal standard

液相色譜串聯質譜分析  
LC-MS/MS analysis

報告限 Reporting limit = 0.01 µg/kg

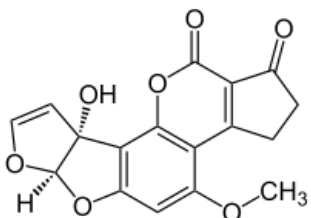
應對每批次的親和柱進行質量驗證  
Performance should be checked  
for each batch of columns



# 真菌毒素檢測 Testing of mycotoxins

## 黃曲霉毒素 Aflatoxins

### Aflatoxin M1



芝士 Cheese

25 g 樣品  
25 g sample



- Add 100 mL dichloromethane and blend for 3 minutes
- Filter, and remove the solvent by rotary evaporator to just dryness
- Reconstituting the residue by dissolving in 5 mL of methanol, 10 mL of heptane and then 30 mL of water by order, and vortex
- Centrifuge at 10000 rpm for 5 minutes
- Collect the lower aqueous layer, Use separating funnel if necessary.
- Further extract the remaining organic layer by 10 mL water,
- Collect the lower aqueous layer.

- 加入 100 mL 二氯甲烷並用均質器混勻 3 分鐘
- 過濾，並用旋轉蒸發溶液
- 依次用 5 mL 甲醇、10 mL 庚烷和 30 mL 水溶解殘留物，然後渦旋均勻
- 離心 10000 rpm 5 分鐘
- 將全部液體移至分液漏斗，收集下層水層。
- 進一步用 10 mL 水 萃取剩餘的有機層
- 收集所有下層水。

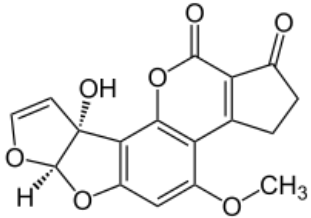




# 真菌毒素檢測 Testing of mycotoxins

## 黃曲霉毒素 Aflatoxins

### Aflatoxin M1



奶 Fluid milk

50 g 樣品  
50 g sample

奶粉, 嬰幼兒配方奶粉

Milk powder, Infant Formula, Follow-up Formula

25 g 樣品  
25 g sample

加入同位素內標  
Spike isotope labelled internal standard

Add 80 mL hot water  
and mix

加入80 mL 熱水  
並攪均

- Centrifuge at 10000 rpm for 10 minutes at about 4°C to defat
- Remove the fat (top) layer
- Proceed to clean-up by IAC with all sample solution.

- 在4°C 中離心 10000 rpm 10 分鐘 以脫脂
- 去除脂肪 (頂層) 層
- 用所有樣品溶液進行IAC萃取

Immunoaffinity SPE Extraction

免疫親和固相萃取

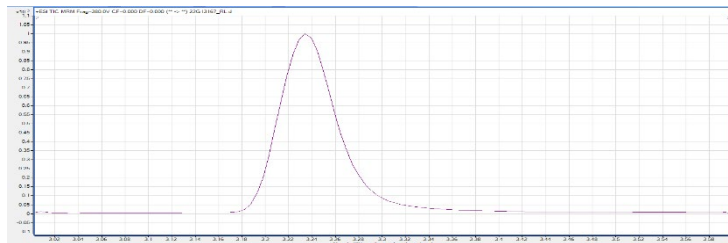
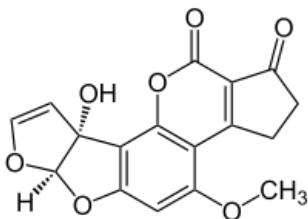
報告限 Reporting limit = 0.01 µg/kg

液相色譜串聯質譜分析  
LC-MS/MS analysis

# 真菌毒素檢測 Testing of mycotoxins

## 黃曲霉毒素 Aflatoxins

### Aflatoxin M1



添加了0.01 ppb 嬰兒配方奶粉的液相串聯質譜色譜  
LC-MS/MS Chromatograms spike at 0.01 ppb (infant formula)



## 液相色譜串聯質譜分析 LC-MS/MS analysis

HPLC system :Agilent 1260 HPLC system or equivalent  
Injection volume :5 µL  
Flow rate :500 µL/min.  
Column : Synergi Polar-RP 80Å, 100 × 2.00mm, 4 micron), or equivalent  
Column temperature :40°C  
Mobile Phase : Solvent A: 0.5 mM ammonium formate and 2 mM formic acid in water  
Solvent B: Acetonitrile

Time, min	Solvent A, % v/v	Solvent B, % v/v
0.00	80	20
2.00	10	90
2.01	80	20
7.00	80	20

MS/MS system : Agilent 6490 triple quadrupole mass spectrometer  
Ionization mode : Electrospray positive ionization  
CUR: : 10  
GAS1: : 25  
GAS 2: : 20  
Spray voltage: : 5500 V  
Source Temperature : 450°C

Analyte	Diagnostic ions (MRM)*				
	Precursor ion (m/z)	Fragment ion (m/z)	Dwell time (ms)	Frag Volt (V)	CE (V)
Aflatoxin M <sub>1</sub>	329	229	100	380	28
		273*	100	380	24
IS	346	288	100	200	4

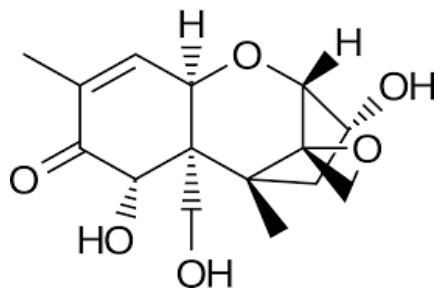
\* Transitions with 100% relative abundance are the base peaks.





# 真菌毒素檢測 Testing of mycotoxins

## 脫氧雪腐鐮刀菌烯醇 Deoxynivalenol



5 g 樣品  
5 g sample

+

加入同位素內標  
Spike isotope labelled internal standard

U-[<sup>13</sup>C<sub>15</sub>]-Deoxynivalenol

- + 30 mL 80% acetonitrile
- Shake 15 mins,
- Homogenize 3 mins
- Centrifuge 10000 rpm 10 mins, filter

- + 30 mL 80% 乙腈
- 振蕩 15分鐘
- 均質 3分鐘
- 離心 10000 rpm 10 分鐘,過濾

### SPE Extraction

- Load 4 mL solution
- Collect 2 mL of eluate

### 固相萃取

- 取 4 mL 溶液至固相淨化柱
- 收集 2 mL 淨化液

- Evaporate to dryness at 50°C under stream of nitrogen
- Reconstitute in 1.0 mL 80% MeOH

- 用氮氣在50°C吹乾
- 加 1 mL 80% 甲醇溶解殘留物

液相色譜串聯質譜分析  
LC-MS/MS analysis

報告限 Reporting limit = 40 µg/kg

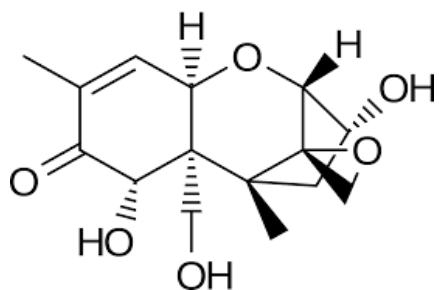


## 真菌毒素檢測

## Testing of mycotoxins

脱氧雪腐镰刀菌烯醇

# Deoxynivalenol



# 液相色譜串聯質譜分析

## LC-MS/MS analysis

HPLC system: Dionex Ultimate 3000 UPLC system or equivalent  
Column: Kinetex C<sub>18</sub> 100 × 3 mm, 100Å, 2.6 µm, Phenomenex  
Injection Volume: 5 µL  
Column Temperature: 35 °C  
Sample Compartment: 10 °C  
Mobile Phase:  
Solvent A: 5 mM ammonium formate with 0.1% formic acid in water  
Solvent B: 5 mM ammonium formate with 0.1% formic acid in methanol

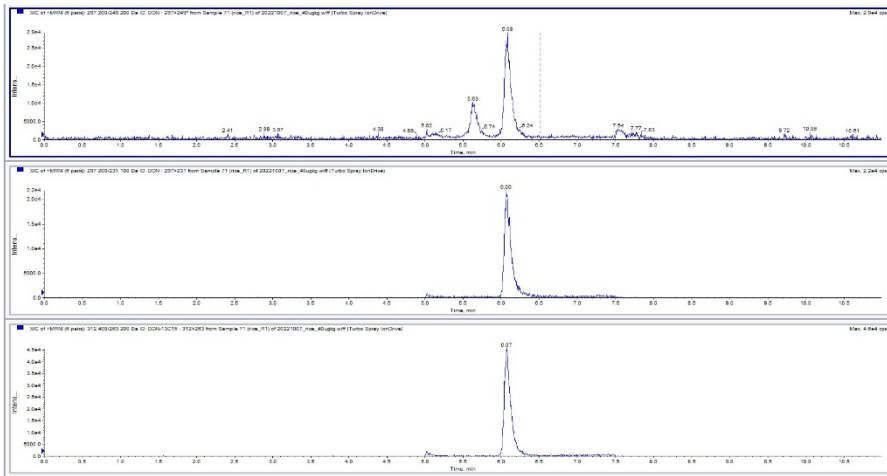
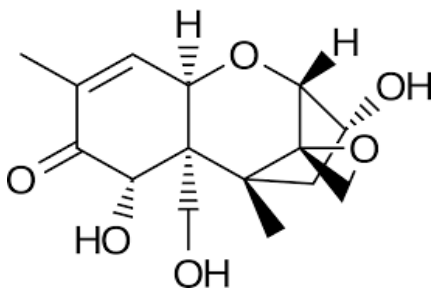
Time, min.	Flow rate, mL/ min	Solvent A, % v/v	Solvent B, % v/v
0.0	0.2	90	10
0.5	0.2	90	10
9.0	0.2	10	90
10.0	0.2	10	90
10.1	0.2	90	10
11.0	0.2	90	10



# 真菌毒素檢測

## Testing of mycotoxins

脫氧雪腐鐮刀菌烯醇  
Deoxynivalenol



## 液相色譜串聯質譜分析 LC-MS/MS analysis

MS/MS System	:	AB SCIEX Triple Quad 6500
Ionization mode	:	Electrospray Positive Ionization
Curtain gas	:	20 psi
Collision gas	:	10 mTorr
Ion Spray voltage	:	5000 V
Temperature	:	50 °C
Ion Source Gas 1	:	75 psi
Ion Source Gas 2	:	30 psi

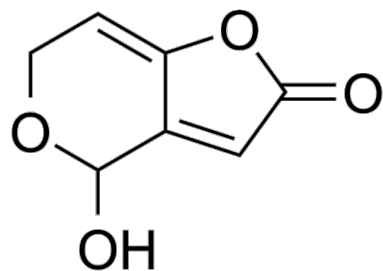
Analyte	Q1	Q3	Dwell time, ms	DP	EP	CE	CXP
DON	297	249*	50	40	10	20	15
	297	231	50	40	10	20	15
	297	203	50	40	10	16	15
<sup>13</sup> C <sub>15</sub> - DON	312	263*	50	40	10	16	15
	312	245	50	40	10	16	15
	312	216	50	40	10	21	15

報告限 Reporting limit = 40 µg/kg

添加了40 ppb 嬰兒純米糊液相串聯質譜色譜  
LC-MS/MS Chromatograms of a rice-based baby cereal spike at 40 ppb

# 真菌毒素檢測 Testing of mycotoxins

棒曲霉素  
Patulin



澄清果汁  
Clear Juice  
無需處理  
No pretreatment

Cloudy Juice 混濁果汁

- 20 g sample
- 20 g 樣品

Apple puree 蘋果果泥

- 20 g 樣品 + 10 mL 水
- 20 g sample + 10 mL water

- + 10 drops of pectinase enzyme
- Leave overnight / 2 hrs @40°C
- Centrifuge 5 mins @3000 rpm

- + 10 滴果膠酶
- 放置過夜/2 小時 @40°C
- 離心 5 分鐘 @3000 rpm

## HLB SPE Cleanup

- Condition 2 mL MeOH and 2 mL H<sub>2</sub>O
- Load 2 mL sample solution to SPE
- Wash the SPE with 2 mL of 1% NaHCO<sub>3</sub> and 2 mL of 1% AcOH. Dry the cartridge
- Eluate the SPE with 2 mL 10% Ethyl acetate in diethyl ether.
- Remove the solvent and reconstitute with 0.5 mL of 2% ACN

## HLB固相萃取淨化

- 分別用 2mL 甲醇和 2 mL 水預淋洗固相萃取柱
- 將 2 mL 樣品溶液轉移至固相萃取柱中
- 次加入 2mL 的 1% 碳酸氫鈉溶液、2mL 1% 醋酸淋洗, 抽乾固相萃取柱
- 加入 2 mL 10% 含乙酸乙酯的二乙醚溶液洗脫, 收集洗脫
- 吹乾並用 0.5 mL 2% 乙腈溶解殘留物

報告限 Reporting limit = 10 µg/kg

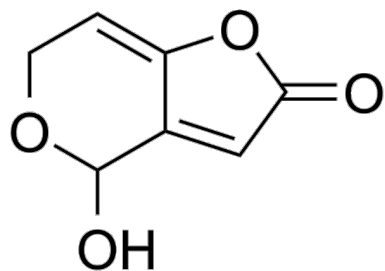
高效液相色譜測定  
HPLC analysis





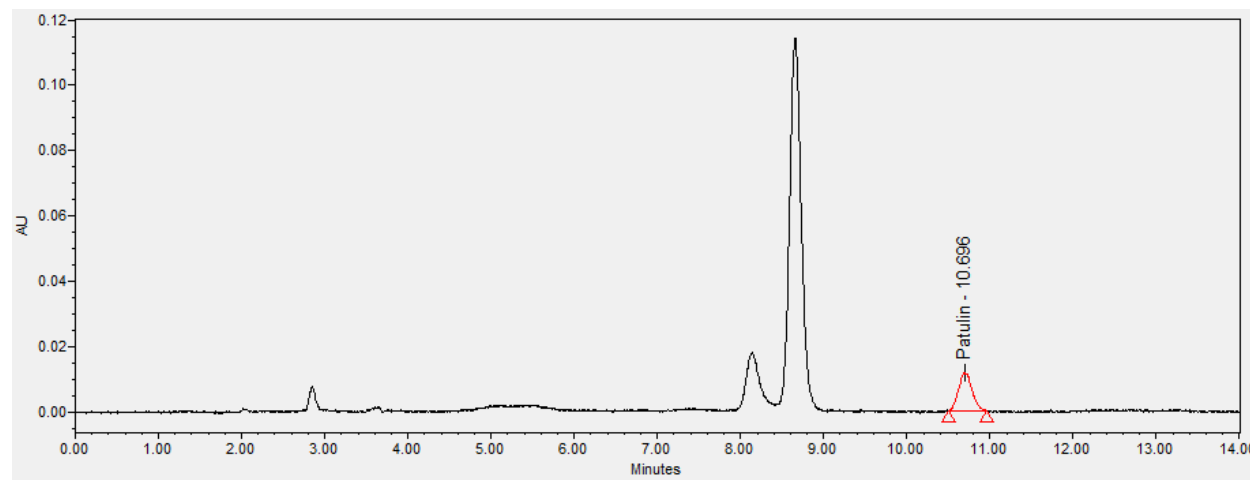
# 真菌毒素檢測 Testing of mycotoxins

## 棒曲霉素 Patulin



## 高效液相色譜測定 HPLC analysis

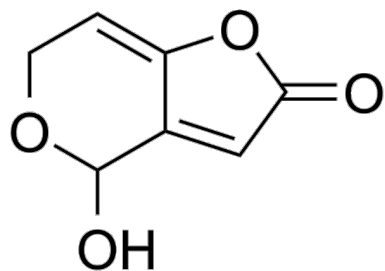
Gradient pump	:	Waters model 600E
Autosampler	:	Waters model 717 <sup>plus</sup>
Detector	:	Waters Photodiode Array Detector model 2475
Column	:	Supelcosil LC-18 DB column, 5 µm, 4.6 mm x 250 mm
Wavelength	:	275 nm (305 nm, if interference peak is suspected)
Injection volume	:	50 µL
Mobile phase	:	THF in water (0.8:100), isocratic





# 真菌毒素檢測 Testing of mycotoxins

棒曲霉素  
Patulin



澄清果汁  
Clear Juice  
無需處理  
No pretreatment

Cloudy Juice 混濁果汁

- 20 g sample
- 20 g 樣品

Apple puree 蘋果果泥

- 20 g 樣品 + 10 mL 水
- 20 g sample + 10 mL water

- + 10 drops of pectinase enzyme
- Leave overnight / 2 hrs @40°C
- Centrifuge 5 mins @3000 rpm

- + 10 滴果膠酶
- 放置過夜/2 小時 @40°C
- 離心 5 分鐘 @3000 rpm

## HLB SPE Cleanup

- Condition 2 mL MeOH and 2 mL H<sub>2</sub>O
- Load 2 mL sample solution to SPE
- Wash the SPE with 2 mL of 1% NaHCO<sub>3</sub> and 2 mL of 1% AcOH. Dry the cartridge
- Eluate the SPE with 2 mL 10% Ethyl acetate in diethyl ether.
- Remove the solvent and reconstitute with 0.5 mL of 2% ACN@pH4

## HLB固相萃取淨化

- 分別用 2mL 甲醇和 2 mL 水預淋洗固相萃取柱
- 將 2 mL 樣品溶液轉移至固相萃取柱中
- 次加入 2mL 的 1% 碳酸氫鈉溶液、2mL 1% 醋酸淋洗, 抽乾固相萃取柱
- 加入 2 mL 10% 含乙酸乙酯的二乙醚溶液洗脫, 收集洗脫
- 吹乾並用 0.5 mL 2% 乙腈 (pH 4) 溶解殘留物

報告限 Reporting limit = 10 µg/kg

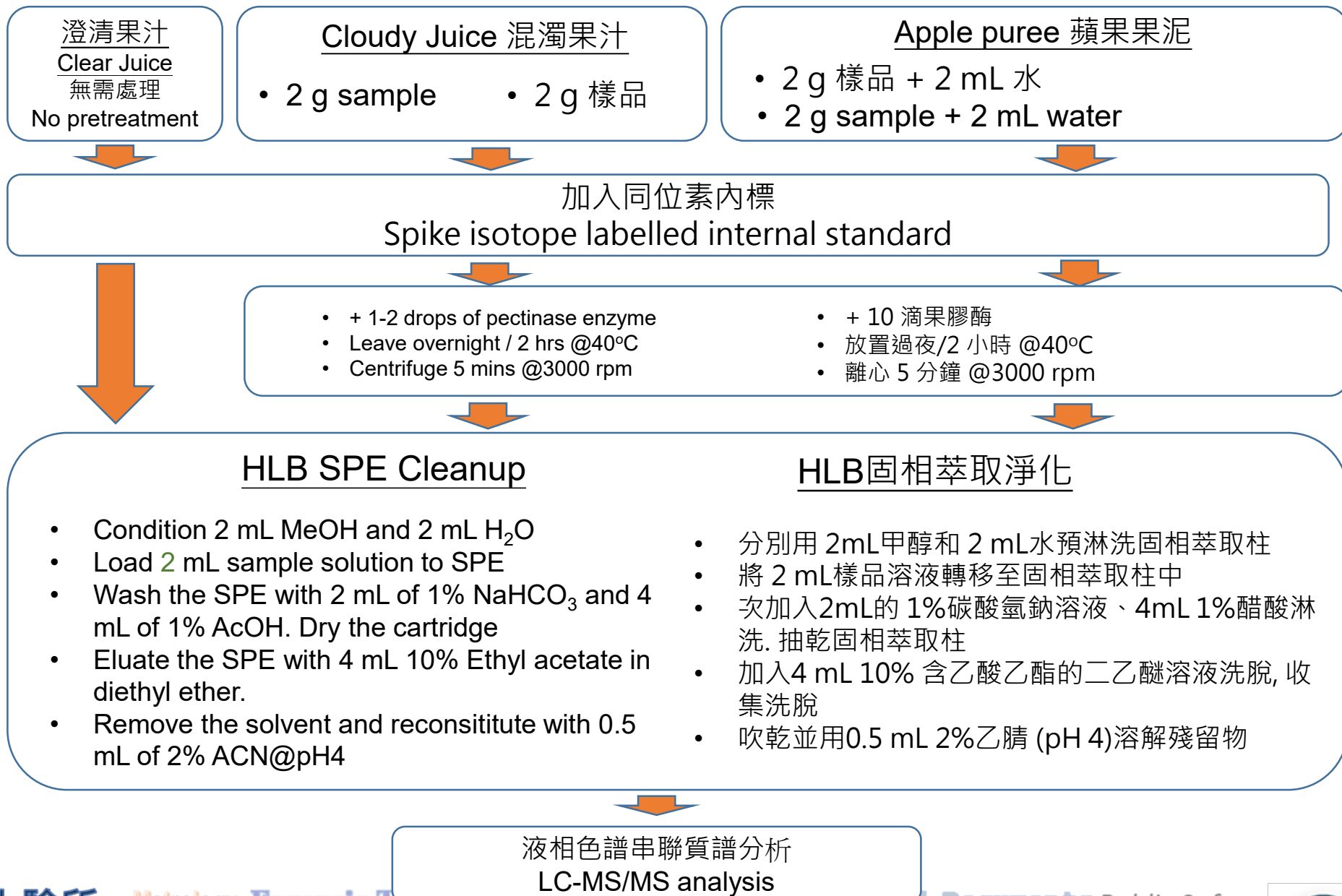
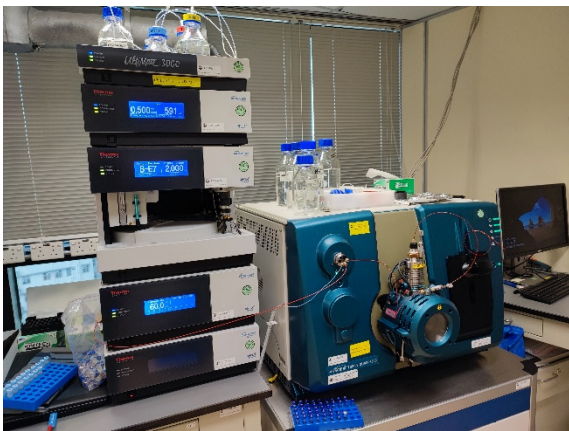
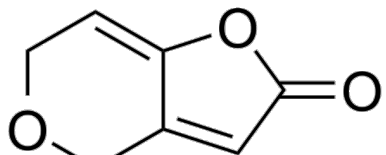
高效液相色譜測定  
HPLC analysis



## 真菌毒素檢測

## Testing of mycotoxins

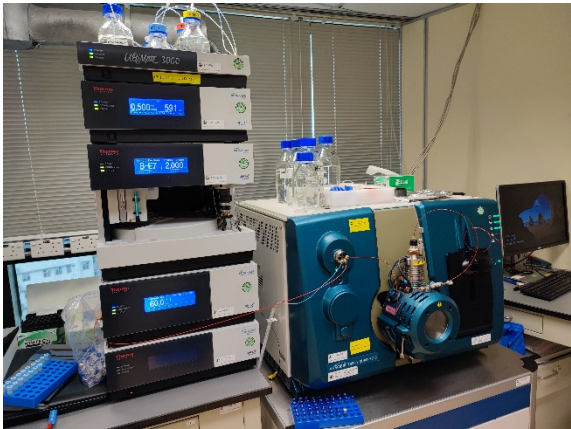
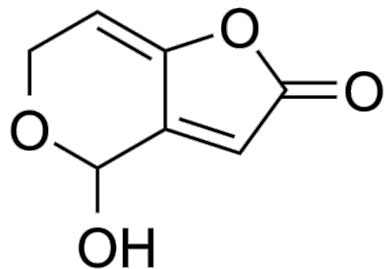
## 棒曲霉素



真菌毒素檢測  
Testing of mycotoxins

液相色譜串聯質譜分析  
LC-MS/MS analysis

棒曲霉素  
Patulin



Injection Volume : 2 µL  
Flow Rate : 250 µL/min  
Column : Scherzo SM-C18, (150 mm × 2 mm, 3 µm), Imtakt  
Mobile Phase : Solvent A: 5 mM ammonium acetate with 3.5 mM acetic acid in water  
Solvent B: Methanol  
Column Temp: 40 °C

Time, min	Solvent A, %v/v	Solvent B, %v/v
0.00	80	20
4.00	80	20
4.01	0	100
5.00	0	100
5.01	100	0
6.00	100	0
6.01	80	20
12.00	80	20

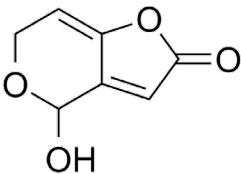


# 真菌毒素檢測 Testing of mycotoxins

## 液相色譜串聯質譜分析 LC-MS/MS analysis

棒曲霉素

Patulin



MS/MS system: Agilent 6490 triple quadrupole MS

Ionization mode: Electrospray negative ionization

Gas Temp: 290 °C

Gas Flow: 20 L/min

Nebulizer: 60 psi

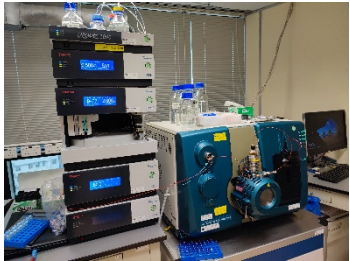
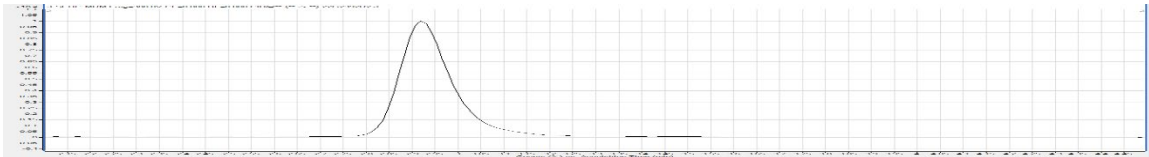
Sheath Gas Heater: 370 °C

Sheath Gas Flow: 12 L/min

Capillary Voltage: -5000 V

Nozzle Voltage: 0

Analyte	Diagnostic ion (MRM)				
	Precursor ion (m/z)	Product ion (m/z)	Dwell time (sec)	Frag Volt (Volt)	CE (V)
Patulin	153	109*	100	380	4
	153	81	400	380	6
Patulin- <sup>13</sup> C <sub>7</sub>	160	115	200	380	2



MS/MS system : AB SCIEX 5500 / MS

Ionization mode : Electrospray negative ionization

Temperature (TEM) : 450 °C

Curtain Gas (CUR) : 20 psi

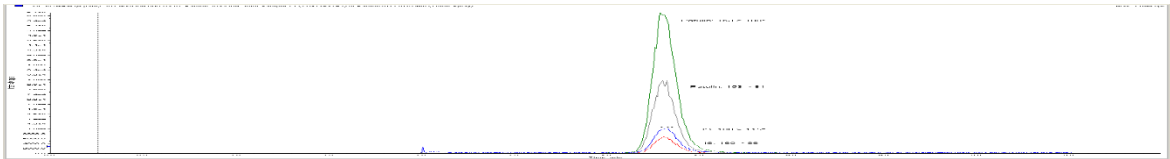
Collision Gas (CAD) : High

Ion Spray Voltage (IS) : -4500 V

Ion Source Gas 1 (GS1) : 50 psi

Ion Source Gas 2 (GS2) : 50 psi

	Diagnostic ions (MRM)						
	Precursor ion (m/z)	Product ion (m/z)	Dwell time (ms)	EP (V)	DP (V)	CE (V)	CXP (V)
Patulin	153	109*	100	-10	-90	-17	-28
		81	100	-10	-90	-13	-35
Patulin- <sup>13</sup> C <sub>7</sub>	160	115	100	-5	-80	-13	-20



真菌毒素檢測  
Testing of mycotoxins

有證標準物質  
Certified Reference Materials

黃曲霉毒素  
Aflatoxins

脫氧雪腐鐮刀菌烯醇  
Deoxynivalenol

棒曲霉毒素  
Patulin

ERM  
Institute for Reference Materials and Measurements

**CERTIFICATE OF ANALYSIS**

ERM®-AC058

AFLATOXIN B2	
Certified value	[µg/g]
Aflatoxin B2	3.80

1) This value was derived from the gravimetric preparation. The value is traceable to the International System of Units (SI).  
2) Estimated expanded uncertainty U with a coverage factor k=2, as defined in the Guide to the Expression of Uncertainty in Measurement (GUM), arising from characterisation as well as from the measurement process.

This certificate is valid for one year after purchase.  
Sales date: 14.02.2022  
The minimum amount of sample to be used is 100 µg.

**NOTE**  
European Reference Material ERM®-AC058 was produced according to the principles laid down in the technical operation agreement between BAM-IRMM-LGC. Info (http://www.erm-crm.org).  
Accepted as an ERM®, Gmel, August 2007

CRM Code: T0X220  
Date of Receipt: 1 Mar 2022  
Concentration: Refer to certificate  
Expiry Date: 14 Feb 2023  
Voucher No.: 01-01-01-01 (2022)

**Uncertainty**  
The certified value(s) and uncertainty(ies) are determined in accordance with EURACHEM/CITAC Guide for "Quantifying Uncertainty in Analytical Measurement, 3rd edition", with a 95% confidence level (k=2). Uncertainty is based on the Total Combined Uncertainty, including uncertainties of characterisation and stability testing. Stability values are based on real evidence opposed to simulation.

The producer certifies that this reference material meets the specification stated in this certificate until the expiry date, provided it is stored unopened at the recommended temperature herein. Product warranties for this reference material are set out in the terms and conditions of purchase.

**CERTIFIED BY** **CERTIFIED ON**  
Ing. M. Streicher 29 Mar 2022

**Signature** **RM Release**

**Logos:** ISO 9001, ISO 17025, COC, Romer Labs Division Holding GmbH, Erber Campus 1, 3131 Getzersdorf, Austria.

0519

ERM  
European Reference Materials

国家标准物质 (NCRM)  
标准物质编号: GBW10174  
Code: CNRM  
国家标准物质资源共享平台  
www.NCRM.org.cn

**标准物质证书**  
Reference Material Certificate

CRM Code: T0X208(a to b)  
Date of Receipt: 9 Jun 2022  
Concentration: Refer to certificate  
Expiry Date: 6 Mar 2024  
Voucher No.: M18-21-0790-ACC

**CERTIFIED REFERENCE MATERIAL CERTIFICATE** ISO 17034

**Certified Reference Material**  
This certificate is designed in accordance with ISO 17034 and ISO Guide 31. This reference material (RM) was designed, produced and verified in accordance with ISO/IEC 17025, ISO 17034 and a registered quality management system ISO 9001.

**Product Name**  
Deoxynivalenol 100 µg/mL in Acetonitrile, Solution of Deoxynivalenol in Acetonitrile

**Product Code** 10000716  
**CAS No.** 51481-10-8  
**Mol. Weight** 296.32  
**Mol. Formula** C15H20O6  
**Volume (ml)** 1

**Lot Number** 1000007890  
**Format** Solution  
**Expiry Date** 6 Mar 2024  
**Storage Temp** 2-8°C

**Chemical Structure:** O=C1C=CC(=C(C=C1)O)O

CERTIFIED	CERTIFIED
Concentration	Expanded Uncertainty (U)
99.98 µg/mL	2.98 µg/mL

**Uncertainty**  
The certified value(s) and uncertainty(ies) are determined in accordance with EURACHEM/CITAC Guide for "Quantifying Uncertainty in Analytical Measurement, 3rd edition", with a 95% confidence level (k=2). Uncertainty is based on the Total Combined Uncertainty, including uncertainties of characterisation and stability testing. Stability values are based on real evidence opposed to simulation.

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**CERTIFIED BY** **CERTIFIED ON**  
Ing. M. Streicher 29 Mar 2022

**Signature** **RM Release**

**Logos:** ISO 9001, ISO 17025, COC, Romer Labs Division Holding GmbH, Erber Campus 1, 3131 Getzersdorf, Austria.

**Certificate of Analysis**  
Aflatoxin M<sub>1</sub> solution

**Description**  
Product ID: CRM46319  
Lot: LRAC6741  
Expiry Date: October 2023  
Manufacturing Date: October 2020  
Storage Conditions: Refrigerate  
Solvent/Matrix: ACETONITRILE

CRM Code: T0X2104  
Date of Receipt: 14 Jun 2021  
Concentration: Refer to certificate  
Expiry Date: Oct 2023  
Voucher No.: C18-21-0180-ACC

**SPExertificate®**  
Certificate of Reference Material

**Catalog Number:** S-4340  
**Description:** Patulin  
**Matrix:** Acetonitrile

**Lot No.** DO200715025  
**Shipment Date:** July 15, 2021  
**Expiration Date:** July 15, 2022

This SPExOrganics® Certified Reference Material, CRM, is intended primarily for use as a calibration standard or quality control standard for organic chromatography instrumentation such as GC, GC-MS, LC, and LC-MS. It can be employed in USEPA, ASTM and other methods relevant to the certified properties listed below.

**Certified Compounds:**

Compound	CAS #	Labeled	Purity	Certified†	Uncertainty
Patulin	149-29-1	1000 µg/mL	98%	1000 µg/mL	± 25 µg/mL

\* - Isomer ratios (when applicable) are an uncertified parameter.

**Final Solution Verification:**  
Final solution integrity verified by Gas Chromatography/Mass Spectrometry. The mass spectrum of each compound was confirmed against the NIST mass spectral database.

† Certified concentration based on gravimetric weights and corrected for the purity of the compound(s) used to prepare the standard. Analytical balance calibration is verified daily with C1 weight set #23-190006 which is registered with Atlantic Scale, and traceable to NIST and NJ Division of Weights and Measures.

This CRM is guaranteed stable and accurate to within the uncertainty listed for the certified value. This includes uncertainty components due to preparation, homogeneity, short term and long term stability. During the stated period of validity, the purchaser will be notified if this product is recalled due to any significant changes in the stability of the solution. For further information, contact the Sales Support Department at crmsales@spexcorp.com.

**Date of Certification:** July 15, 2021  
**Certifying Officer:** Shannon Macleira, Operations Manager

CRM Code: T0X2138(a)  
Date of Receipt: 22 Jul 2021  
Concentration: Refer to certificate  
Expiry Date: 19 Jul 2022  
Voucher No.: C17-21-0076-ACC

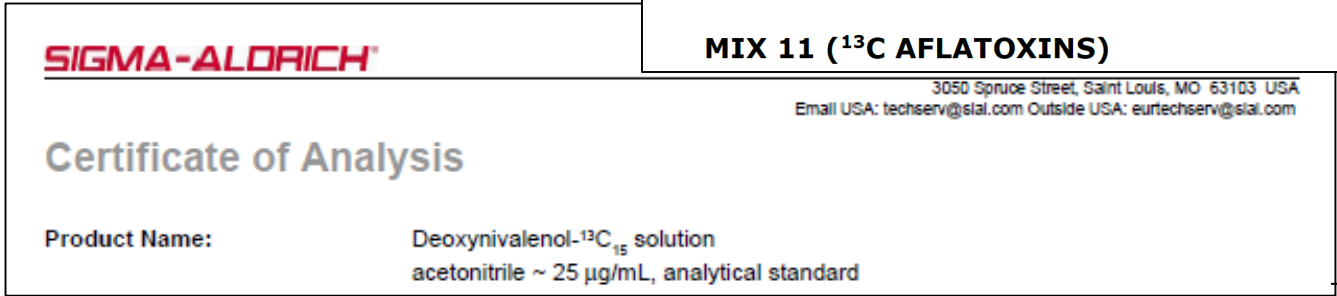
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Rev: 000000



真菌毒素檢測  
Testing of mycotoxins

同位素內標準物質  
Isotope Labelled Internal Standards

黃曲霉毒素  
Aflatoxins



脫氧雪腐鐮刀菌烯醇  
Deoxynivalenol



棒曲霉毒素  
Patulin

