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**INDUSTRY STANDARD OF THE
PEOPLE'S REPUBLIC OF CHINA**

SN/T 3576-2013

**Detection of genetically modified components. Method for
soybean using PCR-DHPLC
(English Translation)**

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Foreword

This standard was drafted in accordance with GB/T 1.1-2009 given rules.

Please note that some of the content of this document may involve patents. Distribution of this document Institutions do not assume the responsibility to identify these patents.

This standard is proposed and managed by the National Certification and Accreditation Administration Committee.

This standard was drafted. People's Republic of China, Shenzhen CIQ, Shenzhen Inspection and Quarantine Science Research Institute, China Inspection Quarantine Science Research Institute, Shenzhen Bo Rui Xiang Hui Biotechnology Co., Ltd., National Center for Nanoscience, Liaoning Academy of Agricultural Sciences.

1 Scope

This standard specifies the PCR-DHPLC detection of genetically modified soybean.

This standard applies to genetically modified soybeans (roundupreadysoybean, RRS) in the detection of genetically modified ingredients.

2 Normative references

The following documents for the application of this document is essential. For dated references, only the dated version suitable for use herein Member. For undated references, the latest edition (including any amendments) applies to this document.

Laboratory use specifications and test methods GB/T 6682 Analysis GB/T 19495.7 GMO detection methods of sampling and sample preparation SN/T 1193 genetic testing laboratory technical requirements Principle 3 Denaturing high-performance liquid

chromatography (denaturing high-performance liquid chromatography, DHPLC) is a simple Single, fast, non-gel method for nucleic acid analysis, samples were analyzed at 50 °C conditions, the number of base pairs in the sample determines the order of elution peak, when over Column acetonitrile concentration increases, the nucleic acid fragment will be from small to large order was eluted according to the relative molecular mass. This standard uses the multiplex PCR the method for genetically modified soybeans containing genetically modified simultaneously amplified PCR products were analyzed by DHPLC, obtained by DHPLC Elution peak marker to determine the relative molecular mass size comparison, it is determined whether or not having a heterologous gene composition and whether the genetically modified soybeans.

4.1 Equipment and Utensils

Tissue grinder, PCR instrument, DHPLC meter, water bath, electronic balance (precision. 1/1000 above), ordinary centrifuge, freezing high Speed centrifuges, refrigerators, cold refrigerator, ice maker, oven temperature, pH meter, pipettes (0.1μL, 0.5μL, 2μL, 10μL, 20μL, 100μL, 200μL, 1000μL), UV-visible spectrophotometer, water machines, autoclaves, PCR tubes (0.2mL), centrifuge tubes (1.5mL, 2.0mL), Tip head (0.1μL ~ 10μL, 5μL ~ 200μL, 100μL ~ 500μL).

4.2 Reagents

CTAB extract, Tris saturated phenol, chloroform, isoamyl alcohol, isopropyl alcohol, 70% ethanol, RNA enzyme (10mg/mL), multiplex PCR the reaction buffer solution (MultiplexPCRMix), TaqDNA polymerase, DHPLC Buffer A. TEAA (0.1mol/L, pH7.0), B Nitrile (0.025%), DHPLC Buffer B. TEAA (0.1mol/L, pH7.0), acetonitrile (25%), DHPLC buffer D. acetonitrile (75%, pH7.0), molecular weight marker (marker).

5 Sampling and sample preparation

According to GB/T 19495.7 performed.