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

SN/T 5200-2020

**Technical specification for identification of pangolin species
(English Translation)**

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Foreword

This document is drafted in accordance with GB/T 1.1-2020.

This document was proposed and managed by the General Administration of Customs of the People's Republic of China.

Drafting organizations of this document. Chinese Academy of Inspection and Quarantine Sciences, Northeast Forestry University.

1 Scope

This document specifies Chinese pangolins, Malay pangolins, Indian pangolins, South African pangolins, giant pangolins, long-tailed pangolins and pangolins.

The morphological characteristics of tree pangolins and other species and the SYBR Green I real-time fluorescent PCR identification method, as well as the sequencing results of PCR products and Sequence analysis and comparison of methods to determine the species of pangolin.

This document applies to Chinese pangolins, Malay pangolins, Indian pangolins, South African pangolins, giant pangolins, long-tailed pangolins and Species identification of tree pangolin and its products.

2 Normative references

The contents of the following documents constitute the indispensable clauses of this document through normative references in the text. Among them, dated quotations Only the version corresponding to that date is applicable to this document; for undated references, the latest version (including all amendments) is applicable Used in this document.

GB/T 6682 Analytical laboratory water specifications and test methods GB 19489 Laboratory Biosafety General Requirements GB/T 27403 Laboratory Quality Control Specification for Food Molecular Biology Testing.

3 Terms, definitions and abbreviations

There are no terms and definitions that need to be defined in this document. The following abbreviations apply to this document.

Cytb. Cytochrome b (cytochrome b) DNA. deoxyribonucleic acid dNTP. deoxy-ribonucleoside triphosphate Real-time PCR. real-time polymerase chain reaction (real-time polymerase chain reaction) SDS. sodium dodecyl sulfate (sodium dodecyl sulfate)

4 Morphological characteristics

Chinese pangolin, Malay pangolin, Indian pangolin, South African pangolin, large pangolin, long-tailed pangolin and adult tree pangolin See Appendix A for typical morphological characteristics.

See Appendix B for the typical morphological characteristics of four pangolin scales. Chinese pangolin, Malay pangolin, tree pangolin and giant pangolin.

Based on the above characteristics, the species of live pangolin or suspected pangolin scale samples can be preliminarily identified, and the precise identification should be further Nucleic acid amplification and sequencing to determine the species.

5.1 Instruments and equipment

5.1.1

Real-time thermal cycler.

5.1.2

Tissue grinder.

5.1.3

Ultra-clean workbench.

5.1.4

Ice maker.

5.1.5

High-speed refrigerated centrifuge.

5.1.6

Water bath.

5.1.7

Desktop small centrifuge.

5.1.8

Conventional refrigerator.

5.1.9

Vortex mixer.

5.1.10

Electrophoresis instrument.

5.1.11

Gel imaging system.

5.1.12

Micro adjustable pipette (10 μ L, 100 μ L, 1,000 μ L) and matching tips.

5.2 Reagents and materials

5.2.1

Anhydrous ethanol.

5.2.2

Proteinase K (20 mmol/ μ L).

5.2.3 50xTAE electrophoresis buffer. For reagent preparation, see C.1 in Appendix C.

5.2.4

Genomic DNA positive control of pangolin. For the preparation of positive materials, see C.2.in Appendix C.

5.2.5

Tirs-Hcl buffer. For reagent preparation, see C.3.in Appendix C.

5.2.6

Physiological saline (0.85% NaCl solution). For reagent preparation, see C.4 in Appendix C.

5.2.7

EDTA solution. see Appendix C C.5 for reagent preparation.

5.2.8

TES buffer. see Appendix C C.6 for reagent preparation.

5.2.9 10% SDS solution. see Appendix C C.7 for reagent preparation.

5.2.10

Sodium chloride.

5.2.11

Trichloromethane.

5.2.12

Isoamyl alcohol.

5.2.13

Glacial acetic acid.

5.2.14

Tris-saturated phenol.

5.2.15 70% ethanol.

5.2.16

SYBR Premix Ex Taq enzyme 1).

5.2.17

Sterilized double-distilled water. should meet the specifications of first-grade water in GB/T 6682.see C.8 in Appendix C.

5.3 Primer pair

Unless otherwise specified, the reagents used in this standard are of analytical grade, and the test water should meet the requirements of GB/T 6682 and GB 19489.

1) TAKARA SYBR Fluorescence Quantitative Kit is an example of a suitable commercially available product. This information is given for the convenience of this standard the user does not mean the approval of this product.