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

SN/T 4278-2015

**Protocol for DNA barcoding identification techniques of
medical insect at frontier ports**

国境口岸医学媒介昆虫DNA条形码鉴定操作规程
(English Translation)

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Foreword

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

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

This standard was drafted. People's Republic of China Guangdong Entry Inspection and Quarantine Bureau, People's Republic of China Hebei Entry-Exit Inspection and Quarantine Bureau.

Introduction

DNA barcoding is a recently developed based on the establishment of a new species identification technology in gene amplification and sequence alignment Surgery, the use of a living body to represent the species standard, there is enough variation, easy and relatively short amplified DNA fragment species Identification. The technology is not the state of development of the species, male and female individuals and morphological integrity or not restrictions, with fast, accurate, low cost Features.

In 2003, Canadian researchers found that PaulHebert mitochondrial cytochrome C oxidase subunit I (COI) gene capable of providing A fast, simple and credible classification method that can accurately distinguish species, and the first time DNA barcoding (DNABarcoding) of concept.

In 2010, co-founded by the 26 countries of the International Consortium for the Barcode of Life (iBOL), we are committed to develop into DNA barcoding General an efficient and accurate way to identify the species, the world has more than 50 countries to participate in this alliance, the purpose is to build a global object Molecular species identification platform. BOLD Barcode Data Systems (Barcodingoflifedatasystem) as an international organization of bar codes Dedicated database and identification platform, screening and voucher specimens DNA barcoding has strict requirements, such as. must have a unique series of samples no details, storage, identification and collection (collection of people,

collecting latitude and longitude location with GPS location information), require gene fragments will it must be greater than 500bp, to provide the use of PCR primer sequences, sequence map, the original sequence or other tracking files. Currently the system is BOLD After more than 200,000 kinds of organisms contain DNA barcode sequence, and are at a very fast pace.

For medical media insect identification relates to border health security, the general use of a length of approximately 658bp for COI gene Standard DNA barcode sequences with BOLD, NCBI database system for comparison.

1 Scope

This standard specifies the medical insect vector DNA barcoding of sample processing, gene amplification, sequence analysis, comparison and identification of junction If it is determined and other operational procedures and norms.

This standard applies to the inspection and quarantine agencies in border crossings using DNA barcoding for adults (including limbs mutilated adults) Identification work and pupae, larvae and eggs and other forms can not rely on direct identification of medical vector insects.

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 19489 General requirements for laboratory biosafety SN/T 1876 medical vectors specimen collection, production and preservation regulations SN/T 2752.4 specification of self-protection of the health and quarantine personnel - Part 4. Laboratory personnel Applications WS/T 230 clinical diagnostic polymerase chain reaction (PCR) technology

3 Terms and Definitions

The following terms and definitions apply to this document.

DNA barcoding DNABarcode to represent the living body of the species, the standard, there is enough variation, and relatively easy to amplify short DNA fragments. DNA barcoding Species identification code has become an important tool in medical vector insects, generally refers to mitochondrial COI (cytochrome C oxidase Standard 658bp fragment of subunit I) on.

DNA barcoding technology DNABarcodingidentificationtechnique A use of DNA barcoding gene fragment species identification and phylogenetic studies of the relationship between

technology, based on gene amplification and on the basis of sequence alignment, it has not species developmental status, male and female individuals and morphological integrity or not to limit, to reduce Experts dependence was not affected by subjective factors, high accuracy, using the Internet can achieve data sharing and so on.

Voucher specimens voucherspecimen Get molecules such as DNA barcoding source data (the parent) specimens. Voucher specimens may be incomplete specimen form, but this standard the molecules of DNA barcode credentials and other data must be a one to one relationship. For the very small sample size (such as mites, etc.), may be the same batch of the same species in a sample of individuals. Save the voucher specimen is very important, it must have a unique number of samples, storage, identification and Mining for more information about sets, etc., in order to facilitate future verification.