

ICS 13.300
CCS G 09

SN

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

SN/T 2497.22-2010

**Test method of import and export dangerous chemicals. Part
22: Determination of DNA T_m**

进出口危险化学品安全试验方法 第22部分：DNA的T_m值测定方法
(English Translation)

Issued on: March 2, 2010

Implemented on: September 16, 2010

Issued by: General Administration of Customs

Table of Contents

Foreword

1 Scope

3 Reagents

3.1 Water

3.2 Sodium chloride - sodium citrate buffer (SSC)

4 Test Procedure

4.1 DNA concentration

4.3 Sample purity

Foreword

SN/T 2497 "Test Methods for Chemical Safety & Exports dangerous" series of standards is divided into 29 parts.

--- Part 1. mammalian liver cells in vivo DNA synthesis (UDS) test;

--- Part 2. plaque-forming cells (PFC) test;

--- Part 3. Large Zhuan reproduction test;

--- Part 4. *Saccharomyces cerevisiae* mitotic recombination test;

--- Part 5. testicular cells UDS test;

--- Part 6. mammalian cells in vitro sister chromatid exchange test;

--- Part 7. mouse ear swelling test;

--- Part 8. popliteal lymph node assay;

--- Part 9. Determination of serum hemolysin test;

Part --- Article 10. T lymphocyte proliferation assay test;

--- Part 11. germline mutation assay;

--- Part 12. single cell gel electrophoresis test;

--- Part 13. fluorescence in situ hybridization;

Part --- Article 14. SDS- polyacrylamide gel electrophoresis;

--- Part 15. PCR-SSCP experiment;

Part --- Article 16. Western-Blot test;

--- Part 17. Behavior of mammalian toxicology tests;

--- Part 18. Detection of DNA adducts;

--- Part 19. NorthernBlot experiment;

--- Part 20. Bradford protein assay;

Part --- Zi 21. agarose gel electrophoresis;

--- Part 22. T_m value determination method of DNA;

--- Part 23. organelle separation experimental methods;

--- Part 24. cellular immunity in vitro assay method;

--- Part 25. humoral immune function tests;

--- Part 26. macrophage function tests;

--- Part 27. Flow cytometry of apoptosis;

--- Part 28. Application shuttle plasmid mutations Inspection;

--- Part 29. Biochemical Oxygen Demand (BOD) was measured.

This part is Part 22 SN/T 2497 series of standards.

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

This section is responsible for drafting unit. People's Republic of China Tianjin Entry-Exit-Entry Inspection and Hunan People's Republic of China Quarantine Bureau.

1 Scope

SN/T 2497 provisions of this part of the test sample and export of dangerous chemicals in the test Test T_m value of DNA, Solution, test procedures and test results.

This section applies to the import and export of dangerous chemicals T_m value measurement test DNA.

Principle 2

DNA in when the temperature rose to a certain value, the hydrogen bonds between the two single-stranded nucleotide began to be opened (DNA begin to denature) Separation, so that the DNA solution is significantly increased absorption in the ultraviolet at a wavelength of 260nm; When the temperature up to a certain value, complete separation of DNA Into single strands, then continue to heat, UV absorption of DNA solution is no longer increases. When DNA thermal denaturation UV absorption maximum increase Thermal denaturation

temperature or melting temperature of the temperature corresponding to half of the DNA is called, i.e. the T_m value.

3.1 Water

This standard test method involves the water are sterile distilled water.

3.2 Sodium chloride - sodium citrate buffer (SSC)

Sodium chloride and trisodium citrate were dissolved in a certain amount of water, such that the sodium chloride concentration of 0.15M solution, the concentration of sodium citrate 0.015M.

4.1 DNA concentration

UV spectrophotometer, determination of DNA sample at 260nm absorbance value A , $A \times 50\mu\text{g/mL}$, the concentration of DNA was calculated.

Sample preparation 4.2 DNA samples taken $60\mu\text{g}$, diluted with 3mL SSC to (0.3 to 0.4 absorbance).

4.3 Sample purity

260nm, 280nm and 230nm absorption at the time by UV spectrophotometer to SSC as a reference solution, the samples were measured 25°C Photometric values A , when 1.75×5 results shall absorbance multiplied by the relative swelling volume when the temperature record and the corresponding absorbance plotted DNA thermal denaturation curve (accurate measurement $V_p T/V_p 25$, corrected for the absorbance of an aqueous solution of $25 \text{ volume deg.}]^\circ\text{C}$), the thermal denaturation midpoint temperature curve corresponding to the T_m value.