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Wool — Colorimetric determination of cystine plus cysteine in hydrolysates

Laine — Détermination colorimétrique de la cystine et de la cystéine dans les hydrolysats

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FOREWORD

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International Standard ISO 2913 was drawn up by Technical Committee ISO/TC 38, *Textiles*, and circulated to the Member Bodies in November 1972.

It has been approved by the Member Bodies of the following countries :

Australia	Germany	Portugal
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The Member Body of the following country expressed disapproval of the document on technical grounds :

France

Wool — Colorimetric determination of cystine plus cysteine in hydrolysates

0 INTRODUCTION

Cystine is the most important amino acid in wool. Its disulphide group is very reactive and is attacked by a wide range of reagents used in wool processing. While acid treatments affect the cystine content only slightly, alkalis, oxidizing and reducing agents, steam and exposure to light and heat cause it to decrease. Since fibre damage is often associated with a decrease in cystine content, a method for its determination is useful for the control and diagnosis of degradation caused by certain agents.

The present method is based on the Folin-Shinohara method for the estimation of cystine in acid hydrolysates of proteins but it should be noted that the cystine content of intact wool is not necessarily the same as that of its hydrolysate. The method is simple to carry out, requires little special apparatus and is suitable for use in industrial laboratories.

The method consists of reductive fission of the disulphide bond of cystine with sodium disulphite, and colorimetric estimation of the cysteine formed, using dodecatungstophosphoric acid. Virgin wool contains a small amount of cysteine and this is included in the cystine plus cysteine content.

1 SCOPE AND FIELD OF APPLICATION

This International Standard specifies a method for the colorimetric determination of cystine plus cysteine in wool hydrolysates.

The method is applicable to all-wool textiles in any form, namely loose fibre, sliver, roving, yarn or cloth. Dyes when present may interfere with the colorimetric measurement. **The method is not applicable to oxidized or reduced wool** because the products of these reactions may undergo side reactions during hydrolysis, thus leading to erroneous results.

The method is therefore primarily applicable to the determination of cystine plus cysteine in washed wool, tops, yarn or cloth that have not been reduced or oxidized.

Metal-complexing agents such as EDTA and cyanide should be absent from the wool, since the reaction is copper catalysed.

2 PRINCIPLE

Hydrolysis of the wool in aqueous sulphuric acid. Reaction of the buffered hydrolysate with sodium disulphite and dodecatungstophosphoric acid, resulting in the development of a blue coloration, the intensity of which is proportional to the concentration of cystine plus cysteine in the hydrolysate. Colorimetric determination of the optical density of the blue solution and calculation of the sum of cystine plus cysteine.

NOTE — The following qualitative test, applicable to undyed wool or a hydrolysate (with dyed wool the hydrolysate must be used), can be used to determine if the wool to be tested contains a significantly higher than normal amount of cysteine.

a) Undyed wool: Immerse the sample in 1 % sodium pentacyanonitrosylferrate solution until thoroughly wetted, remove, dry between filter papers and expose to ammonia fumes. A red-violet coloration indicates cysteine residues.

b) Hydrolysate: Transfer 2 to 3 ml of hydrolysate to a test-tube, add 2 or 3 drops of sodium pentacyanonitrosylferrate solution and make alkaline with concentrated ammonia. An immediate colour change to red-violet indicates cysteine residues.

3 REAGENTS

During the analysis, use only reagents of recognized analytical reagent grade and only distilled water or water of equivalent purity.

3.1 Sulphuric acid, approximately 6 N solution.

Add 150 ml of concentrated sulphuric acid, ρ 1,84 g/ml, 95 to 98 % (m/m) solution, to 850 ml of water.

3.2 Acetate buffer, pH 5,6.

Dissolve 300 g of sodium acetate (dihydrate), 24 ml of glacial acetic acid and 1 mg of copper(II) sulphate (pentahydrate) in water and make up to 1 l.

3.3 Tungstophosphoric acid reagent.

Dissolve 200 g of sodium tungstate (dihydrate—molybdenum free) in 400 ml of water, then add 100 ml of 85 % phosphoric acid and boil gently under reflux for 1 h. Remove the condenser, add bromine or bromine water drop by drop until a bright yellow to brown colour is obtained. Boil off the excess bromine (about 15 min). Cool, filter into a 1 l measuring flask and make up to volume with water. Store the resultant clear, pale yellow-brown solution in a brown glass bottle.