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MODIFIED STANDARD COLORIMETRIC METHOD

FOR DETERMINATION OF

LIGNIN AND TANNIN IN INDUSTRIAL WATERS

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ABSTRACT

A modified Standard Methods is developed for elimination of the interferences caused by calcium ions which are present in many industrial water or wastewater, such as cooling tower water, boiler wastewater, lime-treated paper and pulp wastewater, etc. According to the "Standard Methods for the Examination of Water and Wastewater", Method 5550 Tannin and Lignin [1], 1 mL Folin phenol reagent and 10 mL carbonate-tartrate reagent are added to 50 mL of clear sample (above 20 ^{o}C) and mixed well. After waiting 30 minutes for color development, a spectrophotometric or photometric reading of the sample is to be made and lignin/tannin concentration determined from a calibrated curve previously prepared. When the industrial water or wastewater contains high concentrations of calcium ions, precipitates ($CaCO_3$) are formed at the same time of the color development., therefore, causing interferences at the time of obtaining instrument readings. This publication introduces a modified Standard Method which adds the extra analytical steps of: (a) filtration of the test water sample with 0.45 micron pore-size membrane filter immediately after the completion of the color development period, so accurate instrument readings can be obtained; (b) preparation of calibration curves when using instrument; and (c) preparation of Standard Nessler tubes for color comparison in case an instrument is not available.

Since the analytical procedures introduced in this publication is meant to be a modified "Standard Method", the authors include the exact words in original "Standard Methods 5550 Tannin and Lignin" [1] , but add the authors' recommended extra analytical procedures for the convenience of the readers.

KEYWORDS

Analytical Methods, Tannin, Lignin, Modified Standard Methods, Colorimetric Method, Spectrophotometer, Photometer, Nessler Tubes, Calcium Ion Interferences, Membrane Filtration, Cooling Tower Water, Boiler Water, Industrial Waters

ACRONYM

APHA	American Public Health Association
AWWA:	American Water Works Association
LIWT	Lenox Institute of Water Technology
QC:	Quality control
WEF	Water Environment Federation

MODIFIED STANDARD COLORIMETRIC METHOD FOR DETERMINATION OF LIGNIN AND TANNIN IN INDUSTRIAL WATERS

1. INTRODUCTION

1.1 Environmental Engineering Significance

Lignin is a plant constituent that often is discharged as a waste during the manufacture of paper pulp. [1-3] Another plant constituent, tannin, may enter the water supply through the process of vegetable matter degradation or through the wastes of the tanning industry. [4-7] Tannin also is applied in the so-called internal treatment of boiler waters, where it reduces scale formation by causing the production of a more easily handled sludge. [4-5] Historical development of this analytical method for lignin and tannin determination can be found from the literature [8-11].

1.2 Summary

A modified Standard Methods is developed for elimination of the interferences caused by calcium ions which are present in many industrial water or wastewater, such as cooling tower water, boiler wastewater, lime-treated paper and pulp wastewater, etc. According to the "Standard Methods for the Examination of Water and Wastewater", Method 5550 Tannin and Lignin [1], 1 mL Folin phenol reagent and 10 mL carbonate-tartrate reagent are added to 50 mL of clear sample (above 20 ^{o}C) and mixed well. After waiting 30 minutes for color development, a spectrophotometric or photometric reading of the sample is to be made and lignin/tannin concentration determined from a calibrated curve previously prepared. When the industrial water or wastewater contains high concentrations of calcium ions, precipitates ($CaCO_3$) are formed at the same time of the color development., therefore, causing interferences at the time of obtaining instrument readings. This publication introduces a modified Standard Method which adds the extra analytical steps of: (a) filtration of the test water sample with 0.45 micron pore-size membrane filter immediately after the completion of the color development period, so accurate instrument readings can be obtained; (b) preparation of calibration curves when using instrument; and (c) preparation of Standard Nessler tubes for color comparison in case an instrument is not available.

Since the analytical procedures introduced in this publication is meant to be a modified "Standard Method", the authors includes the exact words in original "Standard Methods 5550 Tannin and Lignin" [1] , but add the authors' recommended extra analytical procedures for the convenience of the readers.

7.2 Chemical Reagent Addition, Chemical Reactions and Color Development

Add in rapid succession 1 mL Folin phenol reagent and 10 mL carbonate-tartrate reagent. Allow 30 min for chemical reactions and color development.

2. PRINCIPLE OF THE STANDARD COLORIMETRIC METHOD

Both lignin and tannin contain aromatic hydroxyl groups that react with Folin phenol reagent (tungstophosphoric and molybdophosphoric acids) to form a blue color suitable for estimation of concentrations up to at least 9 mg/L. However, the reaction is not specific for lignin or tannin, nor for compounds containing aromatic hydroxyl groups, inasmuch as many other reducing materials, both organic and inorganic, respond similarly. [1, 8-11]

3. APPLICABILITY OF THE STANDARD COLORIMETRIC METHOD

This method is generally suitable for the analysis of any organic chemical that will react with Folin phenol reagent to form measurable blue color at the concentration of interest. However, many compounds are reactive and each yields a different molar extinction coefficient (color intensity). Hence, the analyst must demonstrate conclusively the absence of interfering substances.

4. INTERFERENCES OF THE STANDARD COLORIMETRIC METHOD

Any substance able to reduce Folin phenol reagent will produce a false positive response. Organic chemicals known to interfere include hydroxylated aromatics, proteins, humic substances, nucleic acid bases, fructose, and amines. Inorganic substances known to interfere include iron (II), manganese, nitrite cyanide, bisulfite, sulfite, sulfide, hydrazine, and hydroxylamine hydrochloride. Both 2 mg ferrous iron/L and 125 mg sodium sulfite/L individually produce a color equivalent to 1 mg tannic acid/L. [1]

4.1 Minimum Detectable Concentrations

Approximately 0.025 mg/L for phenol and tannic acid and 0.1 mg/L for lignin with a 1-cm-path-length spectrophotometer.

4.2 Quality Control (QC)

The QC practices considered to be an integral part of each method are summarized in Table 5020:1 of the Standard Methods [1]

5. ANALYTICAL APPARATUS

One of the following colorimetric equipment is required:

5.1 Spectrophotometer

A spectrophotometer is used at 700 nm. A light path of 1 cm or longer yields satisfactory results.

5.2 Filter Photometer

A filter photometer is provided with a red filter exhibiting maximum transmittance in the wavelength range of 600 to 700 nm. Sensitivity improves with increasing wavelength. A light path of 1 cm or longer yields satisfactory results.

5.3 Nessler Tubes

They are matched, 100-mL, tall form, marked at 50-mL volume. [1]

6. ANALYTICAL REAGENTS

6.1. Folin Phenol Reagent

Transfer 100 g sodium tungstate, Na_2WO_4 • 2H2O, and 25 g sodium molybdate, Na_2MoO_4 • $2H_2O$, together with 700 mL distilled water, to a 2000-mL flat-bottom boiling flask. Add 50 mL 85% H_3PO_4 and 100 mL cone HC1. Connect to a reflux condenser and boil gently for 10 h. Add 150 g Li_2SO_4, 50 mL distilled water, and a few drops of liquid bromine. Boil without condenser for 15 min to remove excess bromine. Cool to 25°C, dilute to 1 L, and filter. Store finished reagent, which should have no greenish tint, in a tightly stoppered bottle to protect against reduction by air-borne dust and organic materials.

Alternatively, purchase commercially prepared Folin phenol reagent and use before the recommended expiration date.

6.2 Carbonate-Tartrate Reagent

Dissolve 200 g Na_2CO_3 and 12 g sodium tartrate, $Na_2C_4H_4O_6 \cdot 2H_2O$, in 750 mL hot distilled water, cool to 20°C, and dilute to 1 L.

6.3 Stock Solution

The nature of the substance present in the sample dictates the choice of chemical used to prepare the standard, because each substance produces a different color intensity. Weigh 1.000 g tannic acid, tannin, lignin, or other compound being used for boiler water treatment or known to be a contaminant of the water sample. Dissolve in distilled water and dilute to 1000 mL. If the identity of the compound in the water sample is not known, use phenol and report results as “substances reducing Folin phenol reagent” in mg phenol/L. Interpret such results with caution.

Note that tannin and lignin are not individual chemical species of known molecular weight and structure; rather, they are substances containing a spectrum of chemicals of different molecular weights.

Their chemical properties depend on source and method of isolation. If a particular substance is being added to the water, use it to prepare the stock solution.

6.4 Standard Solutions Preparation

Dilute stock solution with distilled water to desired range. Prepare a minimum of 3 standards bracketing expected sample concentration range. [1]

7. ANALYTICAL PROCEDURE

7.1 Sample Storage and Preparation

Bring 50-mL portions of clear sample and standards to a temperature above 20°C and maintain within a ±2°C range.

7.2 Chemical Reagent Addition, Chemical Reactions and Color Development

Add in rapid succession 1 mL Folin phenol reagent and 10 mL carbonate-tartrate reagent. Allow 30 min for chemical reactions and color development.

7.3. Filtration After Color Development

Filter the water sample from Step 7.2 with 0.45 micron pore-size membrane filter right after the completion of the color development period.

7.4 Standard Solution Calibration Curves and/or Standard Solution Nessler Tubes Preparation

7.4.1 Calibration Curves Preparation

Follow the analytical procedures 7.1 to 7.3 using the Standard Solutions prepared in Section 6.4 and using spectrophotometer or photometer for plotting Standard Solution Calibration Curves for lignin or tannin, depending on the standard solution and the target contaminant. Use the following guide for instrumental measurement at a wavelength of 700 nm:

7.4.1.1. Tannic Acid in 61-mL Final Volume

1. Light path 1 cm if tannic acid is in the range of 50-600 ug
2. Light path 5 cm if tannic acid is in the range of 10-150 ug

7.4.1.2. Lignin in 61-mL Final Volume

1. Light path 1 cm if lignin is in the range of 100-1500 ug
2. Light path 5 cm if lignin is in the range of 30-400 ug

7.4.2. Standard Nessler Tubes Preparation

Follow the analytical procedures 7.1 to 7.3 using the Standard Solutions prepared in Section 6.4 and using Nessler Tubes for preparation of a set of Standard Nessler Tubes with known lignin or tannin concentrations.

7.5 Final Lignin or Tannin Concentration Determination

7.5.1. Spectrophotometer or Photometer Application

Make spectrophotometric readings or photometric readings against a reagent blank prepared at the same time, and determine the concentration in accordance with the calibration curves prepared in Procedure Section 7.4.1. Report results in mg/L of the compound known to be present or as “substances reducing Folin phenol reagent” in mg phenol/L.

When using this Modified Standard Methods for regulatory compliance, use spectrophotometric or photometric detection.

7.5.2 Nessler Tube Application

Compare visually against the lignin or tannin Standard Nessler Tubes simultaneously prepared in Section 7.4.2, match the Standard Nessler Tubes and determine the lignin or tannin concentration. Report results in mg/L of the compound known to be present or as “substances reducing Folin phenol reagent” in mg phenol/L.

8. PRECISION AND BIAS

In a single laboratory analyzing seven replicates for phenol at 0.1 mg/L the precision was ±7% and recovery was 107%. [1]

GLOSSARY OF TANNIN AND LIGNIN ANALYSIS

Colorimeter: Nessler tubes and filter photometers are of colorimeters. Both are used for chemical absorption analysis.

Colorimetric method: It is an analytical method involving the development of visible color in standards and water samples after chemical reactions with the specified chemical reagents. Nessler tubes, photometers and spectrophotometers can all be used in colorimetric analysis..

Filter photometer: It is an instrument that uses filters to produce a wide band of wavelengths suitable to colorimetric analysis in which visible color in standards and water samples are developed.

Lignin: It is an organic substance that, with cellulose, forms the chief part of woody tissue. Lignin is plant constituent that often is produced during the manufacture of paper and pulp. Both tannin and lignin contain aromatic hydroxyl groups, and can be quantitatively measured by the same Standard Methods 5550 Tannin and Lignin.

Nessler Tubes: They are identical long glass tubes each with a flat bottom, and each holding about 50 mL capacity for comparison of colored water samples against standards by looking directly down the length of the tube. Human eyes serve as the detector in this visual analysis.

Spectrophotometer: It is an instrument that uses a diffraction grating or a prism to control the light wavelengths used for specific analysis with or without development of visible color.

Tannin: Tannin is a colored plant constituent having aromatic hydroxyl groups. Tannin may enter the water supply through the vegetable matter degradation or through the waste discharge of the leather tanning industry. Both tannin and lignin have been applied in the internal boiler water treatment as the scale control chemical.

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APPENDIX:

INTRODUCTION OF THE EDITORS OF ENVIRONMENTAL SCIENCE, TECHNOLOGY, ENGINEERING AND MATHEMATICS (STEM) SERIES

1. Editor Lawrence K. Wang

Editor Lawrence K. Wang has served the society as a professor, inventor, chief engineer, chief editor and public servant (UN, USEPA, New York State) for 50+ years, with experience in entire field of environmental science, technology, engineering and mathematics (STEM). He is a licensed NY-MA-NJ-PA-OH Professional Engineer, a certified NY-MA-RI Laboratory Director, a licensed MA-NY Water Operator, and an OSHA Instructor. He has special passion, and expertise in developing various innovative technologies, educational programs, licensing courses, international projects, academic publications, and humanitarian organizations, all for his dream goal of promoting world peace. He is a retired Acting President/Professor of the Lenox Institute of Water Technology, USA, a Senior Advisor of the United Nations Industrial Development Organization (UNIDO), Vienna, Austria, and a former professor/visiting professor of Rensselaer Polytechnic Institute, Stevens Institute of Technology, University of Illinois, National Cheng-Kung University, Zhejiang University, and Tongji University. Dr. Wang is the author of 750+ papers and 50+ books, and is credited with 29 invention patents. He holds a BSCE degree from National Cheng- Kung University, Taiwan, ROC, a MSCE degree from the University of Missouri, a MS degree from the University of Rhode Island and a PhD degree from Rutgers University, USA. Currently he is the book series editor of CRC Press, Springer Nature Switzerland, Lenox Institute Press, World Scientific Singapore, and John Wiley. Dr. Wang has been a Delegate of the People to People Internatonal Foundation, a Diplomate of the American Academy of Environmental Engineers, a member of ASCE, AIChE, ASPE, WEF, AWWA, CIE and OCEESA, and a recipient of many US and international engineering and science awards.

2. Editor Mu-Hao Sung Wang

Editor Mu-Hao Sung Wang has been an engineer of the New York State Department of Environmental Conservation, an editor of CRC Press, Springer Nature Switzerland, and Lenox Institute Press, and a university professor of the Stevens Institute of Technology, National Cheng-Kung University, and the Lenox Institute of Water Technology. Totally she has been a government official, and an educator in the USA and Taiwan for over 50 years. Dr. Wang is a licensed Professional Engineer, and a Diplomate of the American Academy of Environmental Engineers (AAEE). Her publications have been in the areas of water quality, modeling, environmental sustainability, solid and hazardous waste management, NPDES, flotation technology, industrial waste treatment, and analytical methods. Dr. Wang is the author of over 50 publications and an inventor of 14 US and foreign patents. She holds a BSCE degree from National Cheng-Kung University, Taiwan, ROC, a MS degree from the University of Rhode Island, RI, USA, and a PhD degree from Rutgers University, NJ, USA. She is the Co-Series Editor of the Handbook of Environmental Engineering series (Springer Nature Switzerland), Coeditor of the Advances in Industrial and Hazardous Wastes Treatment series (CRC Press of Taylor & Francis Group) and the Coeditor of the Environmental Science, Technology, Engineering and Mathematics series (Lenox Institute Press). She is a retired member of AWWA, NYWWA, NEWWA, WEF, NEWEA, CIE and OCEESA, and is currently serving as a member of the APHA/AWWA/WEF Standard Methods committee.

3. Editor Yuriy I. Pankivskyi

Editor Yuriy I. Pankivskyi has 25 years of professional experience of scientific research and environmental education. He has expertise in strategic environmental assessment, environmental impact assessment, drinking water treatment, waste waters treatment, water and air pollution control, solid waste management. He works as environmental consulting engineer for industrial enterprises, state administrations of cities and towns of Western Ukraine, communities, private firms and institutions and as researcher, educator for state universities. He is the Associate Professor and Deputy Head of Department of Ecology of Ukrainian National University of Forestry. His research and publications have been in areas of water and air quality control, waste water treatment, environmental sustainability and education, analytical methods, investigations of multifunctional material for optoelectronics and environment testing. Dr. Pankivskyi is author of over 70 scientific publications. He earned his Specialist degree from Lviv State Ivan Franko University (Ukraine), ME degree from Lenox Institute of Water Technology (MA, USA), and his PhD degree from Lviv National Ivan Franko University (Ukraine). He is a member of National Ecological Center of Ukraine (Lviv Department).