

Copper in Soils

Photometric determination with Cuprizon subsequent to acid extraction

Introduction

The determination of copper in soil is important due to its role as an essential plant micronutrient involved in metabolic processes such as photosynthesis, respiration, and protein synthesis.¹ However, elevated copper levels can negatively affect plant growth and productivity, including impacts on germination and physiological processes.² Copper contamination in agricultural soils is common and is associated with the use of fertilizers, pesticides, and copper-based fungicides, making its monitoring important.

A range of analytical methods is available for copper determination, including atomic absorption spectroscopy and inductively coupled plasma techniques. Photometric methods based on complex formation provide an alternative approach for copper analysis following extraction.

Experimental

This application note details the photometric determination of copper in soils, using Glyoxal-bis (2-hydroxyanil), after its extraction.

Method

In an ammoniacal medium copper(II) ions react with cuprizon to form a blue complex that is determined photometrically.

Measuring Range

Spectroquant® Copper Cell Test (1.14553)	Test kit: 0.05–8.00 mg/L Cu
	Method: 1.00–160.0 mg/kg Cu
Spectroquant® Copper Test (1.14767)	Test kit: 0.02–6.00 mg/L Cu
	Method: 0.40–120 mg/kg Cu

Applicable Sample

Soil samples

Reagents, Instruments and Materials

Test/Reagent Kit(s)

For the measurement one of the following Spectroquant® test kits is necessary:

- Spectroquant® Copper Cell Test (1.14553)
- Spectroquant® Copper Test (1.14767)

Instrument(s) & Devices

For the measurement one of the following Spectroquant® photometers is necessary:

- Spectroquant® VIS Spectrophotometer Prove 100 plus (1.73026)
- Spectroquant® UV/VIS Spectrophotometer Prove 300 plus (1.73027)
- Spectroquant® UV/VIS Spectrophotometer Prove 600 plus (1.73028)
- Spectroquant® Colorimeter Move 100 (1.73632)

Note: Also, legacy Spectroquant® instruments are suitable.

Software for Data transfer

- Optional Spectroquant® Prove Connect to LIMS software package (Y.11086) to transfer your data into an existing LIMS system.

Instrument Accessories

- Rectangular cells 10 mm (1.14946)
- Rectangular cells 20 mm (1.14947)
- Rectangular cells 50 mm (1.14944)

Other Reagents and Accessories

- Nitric acid 65% for analysis (**1.00456**)
- Ammonia 25% for analysis (**1.05432**)
- MQuant® pH-indicator strips pH 7.5–14 (**1.09532**)
- Water for analysis (**1.16754**)
- Analytical balance
- Standard laboratory glassware (e.g., glass beakers) and pipettes
- Shaker
- Sieve
- Heating plate
- Folded filter

Analytical Procedure

For more information on the measurement see the packaging insert for the test.

Sample Preparation

- In a 200 mL Erlenmeyer flask place 10 g of air dried and sieved (mesh 2 mm) sample, with 30 mL water for analysis and 10 mL nitric acid 65% and heat for 1 hour.
- After cooling slowly and under swinging add 23 mL ammonia 25%. The pH - value should be 9–10 (check with pH-indicator strips).
- Filter off the insoluble parts and wash the filter 3 times with water for analysis.
- Collect the filtrate and the washings in a 200 mL volumetric flask, fill up to volume with water for analysis and mix well.

Using Cat. No. 1.14553: Procedure and measurement

For more information on the measurement see the packaging insert for the test

Procedure

- Pipette 5.0 mL Pretreated sample into a reaction cell, close the cell, and mix. The pH must be within the range 7.0–9.5. Check with MQuant® pH-indicator strips.
- Add 5 drops Reagent Cu-1K, close the cell tightly, and mix. Hold the bottle vertically while adding the reagent!
- Leave to stand for 5 min (reaction time), then measure the sample in the photometer.

Measurement

- For photometric measurement the cells must be clean. Wipe, if necessary, with a clean dry cloth.
- Measurement of turbid solutions yields false-high readings.
- The pH of the measurement solution must be within the range 7.0–9.5.
- The color of the measurement solution remains stable

for at least 30 min after the end of the reaction time stated above. (After 45 min the measurement value would have diminished by 5%.)

- In the event of copper concentrations exceeding 50 mg/l, other reaction products are formed, and false-low readings are yielded. In such cases it is advisable to conduct a plausibility check of the measurement results by diluting the sample (1:10, 1:100).

Hints for Measurement

- It is recommended to zero the method each new working day. To do this, open the method, either by manually selecting the method or by inserting a barcoded cell. Tap the <Settings> button and select the <ZERO ADJUSTMENT> menu item. After prompting, insert the 16 mm zero cell through the corresponding opening. The zero adjustment is performed automatically. Confirm the performance of the zero-adjustment procedure by clicking on <OK>.
- After the zero has been performed, insert the barcoded Spectroquant® round cell through the corresponding opening, ensuring that the white position mark on the cell is aligned with the positioning mark on the spectrophotometer. The measurement starts automatically.
- Read off the result in mg/L from the display.

Hint: The above written measurement description is only valid for the Spectroquant® Prove (plus) series photometer. If a different instrument is used, please consult the corresponding instrument manual for more details on how to perform the measurement.

Using Cat. No. 1.14767: Procedure and measurement

For more information on the measurement see the packaging insert for the test.

Procedure

- Pipette 5.0 mL Pretreated sample into a test tube.
- Add 1 level green dosing spoon Reagent Cu-1 and shake vigorously until the reagent is completely dissolved.
- The pH must be within the range 7.0–9.5. Check with MQuant® pH-indicator strips.
- Add 5 drops Reagent Cu-2 and mix. Hold the bottle vertically while adding the reagent!
- Leave to stand for 5 min (reaction time), then fill the sample into the cell, and measure in the photometer.

Notes:

- For measurement in the 50 mm cell only the sample volume must be doubled. Alternatively, the semi-microcell (**1.73502**) can be used. It is recommended to measure against an own prepared blank sample (preparation as per measurement sample, but with distilled water instead of sample) to increase the accuracy. Configure the photometer for blank measurement

Measurement

- Certain photometers may require a blank (preparation as per measurement sample, but with distilled water instead of sample).
- For photometric measurement the cells must be clean. Wipe, if necessary, with a clean dry cloth.
- The pH of the measurement solution must be within the range 7.0–9.5.
- The color of the measurement solution remains stable for at least 30 min after the end of the reaction time stated above. (After 45 min the measurement value would have diminished by 5%.)
- In the event of copper concentrations exceeding 50 mg/L, other reaction products are formed, and false-low readings are yielded. In such cases it is advisable to conduct a plausibility check of the measurement results by diluting the sample (1:10, 1:100).

Hints for Measurement

- It is recommended to zero the method for each new working day. To do this, open the method by inserting the barcode, tap the <Settings> button and select the <ZERO ADJUSTMENT> menu item. Fill the same cell which will be used for the sample measurement with distilled water. After prompting, insert the filled rectangular cell into the cell compartment. The zero adjustment is performed automatically. Confirm the performance of the zero-adjustment procedure by clicking on <OK>.
- After the zero adjustment, fill the measurement sample into the same or a matched rectangular cell and insert the cell into the cell compartment. The measurement starts automatically.

- Read off the result in mg/L from the display.

Hint: The above written measurement description is only valid for the Spectroquant® Prove (plus) series photometer. If a different instrument is used, please consult the corresponding instrument manual for more details on how to perform the measurement.

Analytical Quality Assurance

Analytical quality assurance (AQA) is recommended before each measurement series.

To check the photometric measurement system (test reagents, measurement device, handling) and the mode of working, Spectroquant® CombiCheck 90 can be used. Besides a standard solution with 2.00 mg/L Cu²⁺, this article also contains an addition solution for determining sample-dependent interferences (matrix effects).

Sample-dependent interferences (matrix effects) can be determined by means of standard addition or dilution.

To view additional notes, visit

[SigmaAldrich.com/qa-test-kits](https://www.sigmaaldrich.com/qa-test-kits).

Calculation

Copper content in mg/kg Cu = analysis value in mg/L Cu x 20

References

1. Mir, A. R.; Pichtel, J.; Hayat, S. Copper: Uptake, Toxicity and Tolerance in Plants and Management of Cu-Contaminated Soil. *Biometals* **2021**, 34 (4), 737–759. DOI: [10.1007/s10534-021-00306-z](https://doi.org/10.1007/s10534-021-00306-z).
2. Gonzaga, M. I.; Matias, M. I.; Andrade, K. R.; de Jesus, A. N.; da Costa Cunha, G.; de Andrade, R. S.; de Jesus Santos, J. C. Aged Biochar Changed Copper Availability and Distribution among Soil Fractions and Influenced Corn Seed Germination in a Copper-Contaminated Soil. *Chemosphere* **2020**, 240, 124828. [10.1016/j.chemosphere.2019.124828](https://doi.org/10.1016/j.chemosphere.2019.124828).

Featured Products

Description	Cat. No.
Test Kits	
Spectroquant® Copper Cell Test	1.14553
Spectroquant® Copper Test	1.14767
Instruments & Accessories	
Spectroquant® VIS Spectrophotometer Prove 100 plus	1.73026
Rectangular cells 10 mm	1.14946
Rectangular cells 20 mm	1.14947
Rectangular cells 50 mm	1.14944
Reagents & Test Strips	
Nitric acid 65% for analysis	1.00456
Ammonia 25% for analysis	1.05432
MQuant® pH-indicator strips pH 7.5–14	1.09532
Water for analysis	1.16754

Related Products

Description	Cat. No.
Instruments & Software	
Spectroquant® UV/VIS Spectrophotometer Prove 300 plus	1.73027
Spectroquant® UV/VIS Spectrophotometer Prove 600 plus	1.73028
Spectroquant® Colorimeter Move 100	1.73632
Spectroquant® Prove Connect to LIMS software package	Y.11086
Reference Materials	
Spectroquant® CombiCheck 90	1.18700
Reagents, Test Strips, & Accessories	
Nitric acid 65% for analysis EMSURE®	1.00456
Spectroquant® Crack Set 10C	1.14688
Spectroquant® Crack Set 10	1.14687
Empty cells 16 mm with screw caps (25 pcs)	1.14724
Hydrogen peroxide 30% H ₂ O ₂ (Perhydrol®) for analysis EMSURE®	1.07209
MQuant® Copper Test, measuring range 10–300 mg/L Cu	1.10003
MQuant® Universal indicator strips pH 0–14	1.09535
MQuant® pH-indicator strips pH 5.0–10.0	1.09533
Sodium hydroxide solution 1 mol/L Titripur®	1.09137
Sulfuric acid 0.5 mol/L Titripur®	1.09072
Semi-microcells 50 mm (2 pcs)	1.73502

For more information visit us at

SigmaAldrich.com/photometry



To place an order or receive technical assistance:
SigmaAldrich.com/support



For local contact information:
SigmaAldrich.com/offices

MilliporeSigma
400 Summit Drive
Burlington, MA 01803
SigmaAldrich.com

We have built a unique collection of life science brands with unrivalled experience in supporting your scientific advancements.

Millipore® Sigma-Aldrich® Supelco® Milli-Q® SAFC® BioReliance®

© 2026 Merck KGaA, Darmstadt, Germany and/or its affiliates. All Rights Reserved. MilliporeSigma, the vibrant M, BioReliance, Millipore, Milli-Q, SAFC, Sigma-Aldrich, Supelco, Spectroquant, MQuant, EMSURE, and Titripur are trademarks of Merck KGaA, Darmstadt, Germany or its affiliates. All other trademarks are the property of their respective owners. Detailed information on trademarks is available via publicly accessible resources.

MS_AN14527EN Ver. 1.0 71305 09/2026

