

# Soil Analysis by Mehlich 3 Extractant Technique Using the Microlab 600 Diluter for Sample Preparation

## Authors

### ÚKZUZ (Ústřední kontrolní a zkušební ústav zemědělský)

Central Institute for Supervising and Testing in Agriculture  
Hroznová 2, CZ-65606 Brno, Czech Republic

### L. Vlk, M. Horová, R. Krejča; R. Špejra

Chromservis S.R.O., Jakobiho 327, CZ-10900 Praha-10, Petrovice, Czech Republic



## Introduction

Mehlich 3 has been used in the determination of macro and micronutrients in soils and also for the agrochemical testing of agricultural soil in the Czech Republic since 1999. This systematic soil testing is carried out in six-year cycles for the purpose of agrochemical control of available nutrient conditions, soil reaction and liming needs. Agrochemical soil testing is a method of monitoring exchangeable soil reaction, carbon content, liming need and the determination of available phosphorus and potassium. These data were used to create the basic development of fertilizer plans of agricultural crops, monitoring of soil properties and forecasting of fertilizing needs.

The available nutrient content in the soil determined by Mehlich 3 is increased compared to the Mehlich 2 method used before 1999. The phosphorus content is increased by 20%, while potassium and magnesium are increased by 3-4% due to the better displacement of individual elements from the soil sorption complex using the Mehlich 3 method. This data has to be taken into account when comparing results from studies conducted prior to 1999.

Basic soil properties are evaluated separately for arable land, permanent grassland, vineyards, fruit farms and hop gardens. On average about 75,000 soil samples are analyzed annually within Czech National Reference Laboratory (NRL) ÚKZUZ. This means that the Hamilton Advanced Microlab Diluter (p/n ML625-DIL) will perform more than 60,000 dilutions per year in this laboratory.

## Material & Methods

### Mehlich 3 Extractable Elements

The Mehlich 3 method is a weak acid soil extraction procedure that has the advantage of being applicable for a number of elements. The extract is composed of 0.2 M glacial acetic acid, 0.25 M ammonium nitrate, 0.015 M ammonium fluoride, 0.013 M nitric acid, and 0.001 M ethylene diamine tetraacetic acid (EDTA). Mehlich 3 is most commonly used for determination of macronutrients (phosphorous, calcium, magnesium and potassium) and micronutrients (copper, zinc, manganese, and iron). Phosphorus is extracted by a reaction with acetic acid and fluoride compounds. Mehlich 3 has been found to be closely related to other conventional methods of phosphorus-extraction such as Olsen and bicarbonate. Mehlich 3 is highly correlated with plant phosphorus uptake and is therefore considered as a standard method for available phosphorus determination.

### Element analysis

Exchangeable base cations (calcium, magnesium, and potassium) are extracted by ammonium nitrate and nitric acid. Micronutrients (copper, zinc, manganese, and iron) are extracted by ammonium and the chelating agent EDTA. The amounts recovered are linearly related to pentaacetate (DTPA) and 0.1 M HCL methods. FAAS Flame Atomic Absorption Spectroscopy (FAAS) method is used for measuring the micronutrient concentration. Potassium is concentration determined by Flame Atomic Emission Spectroscopy (FAES). Calcium and magnesium concentrations are determined by FAAS in the air-acetylene flame after dilution in the presence of lanthanum. Phosphorus concentration is determined by using the ascorbic-ammonium molybdate method. The intensity of blue coloration is measured by UV-VIS spectrophotometer.

## Reagents

1. Ammonium fluoride – EDTA stock solution is prepared by dissolving 138.9 g of ammonium fluoride (NH<sub>4</sub>F) in 600 mL of deionized water and then adding 73.5 g EDTA dissolve and dilute to 1000 mL.
2. Mehlich-3 extracting solution is prepared by dissolving 200.0 g ammonium nitrate (NH<sub>4</sub>NO<sub>3</sub>) in about 6,000 mL of deionized water. Then add 40.0 mL of the ammonium fluoride-EDTA stock solution and mix well. Next add 115 mL of concentrated glacial acetic acid (CH<sub>3</sub>COOH) and 8.25 mL of concentrated nitric acid (HNO<sub>3</sub>) and finally bring to 10,000 mL final volume. The final pH should be 2.50 ± 0.05.

## Apparatus

1. Hamilton Advanced Microlab 625 Diluter (p/n ML625-DIL) with 10 mL and 500 µL syringes

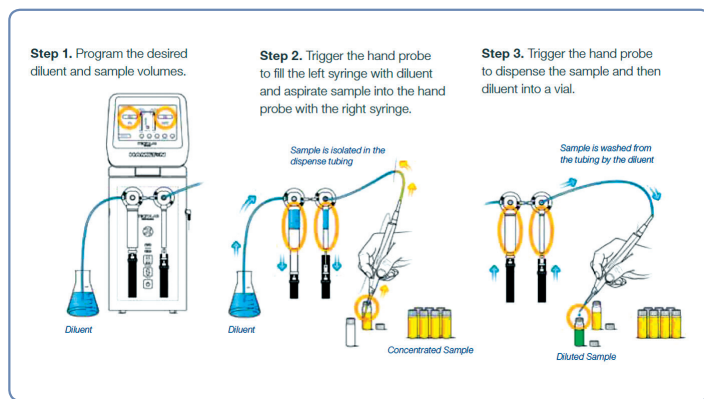


Figure 1: Schematic illustration of the dual diluter method.

2. FAAS Perkin Elmer AA 400 Spectrophotometer for analyzing calcium and magnesium concentrations.
3. Varian 500 CONC, for analyzing phosphorous concentrations.

## Extraction

1. Weigh 10.0 ± 0.05 g of air dried soil (particle size < 2.0 mm) into a 250 mL plastic bottle.
2. Add 100 mL of Mehlich 3 Extractant. Include a method blank and check samples as determined by the lab.
3. Place extraction bottles on rotary shaker (35 ± 5 rpm) for ten minutes.
4. Centrifuge suspension at 3500 rpm for 5 min.

## Results and Discussion

The Advanced Microlab Diluter was used for sample preparation of the soil extracts as described above. The semi-automated dispensing capabilities can be used for accurate and precise dilutions of the extracts for determination of calcium, magnesium and phosphorous. Using this method a soil samples may be analyzed with excellent accuracy and precision with reproducible results compared to manual liquid handling techniques. This conclusion is supported by the data shown in Figure 2. In addition, the Microlab 600 instruments are positive displacement devices that offer unmatched accuracy compared to air displacement systems, such as pipettes where the real value of the dispensed volume often depends on the liquid's physical properties and/or operating conditions.

Product	Diluter Hamilton
Identification	Microlab 625 DIL
T [°C]	22
Density [µL/mg]	1.0032
Date	24.04.2013
Average volume V (µL)	5252.0
Relative accuracy A %	0.04
Standard deviation s (µL)	1.995222197
CV %	0.038004232
Permissible tolerance	1%
Conclusion	dilutor suits

Product	Diluter Hamilton
Identification	Microlab 625 DIL
T [°C]	22
Density [µL/mg]	1.0032
Date	15.10.2012
Average volume V (µL)	5255.6
Relative accuracy A %	0.11
Standard deviation s (µL)	0.791334832
CV %	0.015073044
Permissible tolerance	1%
Conclusion	dilutor suits

Figure 2: Customer test reports - Measured average volumes dispensed by the Microlab 600 for two Mehlich 3 tests (volume was checked with balance, 10 single measurements per test). This data demonstrates the accuracy and precision of the Advanced Microlab 600 Diluter during two different days.

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Web: [www.hamiltoncompany.com](http://www.hamiltoncompany.com)

USA: 800-648-5950

Europe: +41-81-660-60-60

**Hamilton Americas & Pacific Rim**  
4970 Energy Way  
Reno, Nevada 89502 USA  
Tel: +1-775-858-3000  
Fax: +1-775-856-7259  
[sales@hamiltoncompany.com](mailto:sales@hamiltoncompany.com)

**Hamilton Europe, Asia, & Africa**  
Via Crusch 8  
CH-7402 Bonaduz, Switzerland  
Tel: +41-81-660-60-60  
Fax: +41-81-660-60-70  
[contact@hamilton.ch](mailto:contact@hamilton.ch)

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