A Low Cost Miniature Method to Determine Iron Content in Samples Suitable for Small Research Laboratories

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Introduction

Iron toxicity is a serious problem in lowland rice production systems. In its reduced form (Fe^{2+}), it is available to the plant due to flooding. Determination of iron contents in plant or soil samples is relatively laborious and involves expensive equipment such as a high-pressure acid digestion system and an atomic absorption spectroscope. The method requires relatively large amounts of sample material and due to the costs involved only a limited number of samples can be analysed per day. Therefore, this method is in many cases unsuited for laboratories of small research or field stations. In order to reduce sample size as well as costs and equipment requirements we developed an analytical method that uses micro titer plates and a plate reader.

Conclusions

- Iron was successfully determined with the new Micro-Method.
- Na dithionite reduced all Fe III in the sample to Fe II.
- Low pH in acid extracted samples disturbed the color reaction on the plates.
- Hot water extracted only a fraction of the tissue iron content.
- Further research is needed to identify the fraction of tissue iron content extractable by water.

Methodology Development

Standard Method

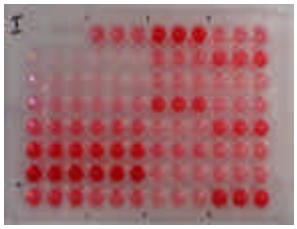
- Atomic Absorption Spectrometry
- 1g sample size
- In 7 mL H_3PO_4 acid
- High pressure acid digestion (180°C, 2h)
- fill up to 100 mL
- Measurement of Fe with AAS

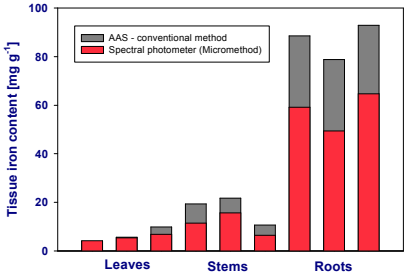
- Trained staff required
- High equipment costs
- Specific laboratory space needed
- Low sample turn-over rates

- High precision level
- Results compatible among labs

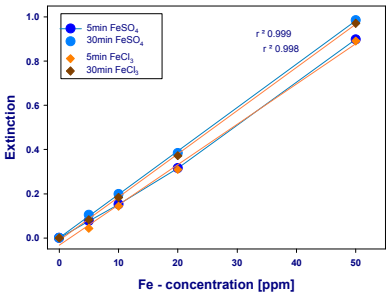
Micro Method

- Spectral-photometry using Micro-titerplates at 490 nm.
- 0.02-0.05 g sample size
- In 5 mL H_2O
- high pressure hot water digestion (autoclave)
- fill up to 10 mL
- each well comprises:
  - 100 µL sample
  - 150 µL 2,2 Di-pyridyl (5 m*M*) (color agent)
  - 50 µL Na-dithionite (50mM) (reducing agent)
- No trained staff required, low equipment costs
- High sample turn-over rate
- Needs different extraction method

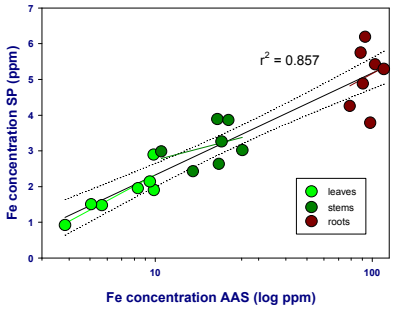




Iron recovery from the same sample when reduced to Fe II varied between 80 – 100 % of the results obtained with AAS depending on the analysed tissue



Using $FeCl_3$ for standards proved the concentration of NDT sufficient to fully reduce FeIII to FeII in the sample



When extracted with hot water instead of acid between 20% (roots) and 95% (leaves) of the tissue iron content were found

After shaking and allowing for a minimum reaction time of 30 min extinction values were constant