

Optimization of Methods for Analysis of Phosphatase, β -Glucosidase and Arylsulfatase in Soils

Guilherme M. Chaer¹, Leonardo V. B. Pazutti²

¹Embrapa Agrobiologia, Seropédica, Brazil; ²Universidade Federal Rural do Rio de Janeiro, Brazil.

Introduction

Because of the readiness of response of microorganisms to changes in soil environment, some hydrolytic soil enzymes have been widely proposed as sensitive indicators of soil quality. However, costs of several enzymatic analyses in soil laboratories are still high compared with the traditional routine soil fertility tests.

Objective

To evaluate the feasibility of modifications in the commonly used methods for the analysis of phosphatase, β -glucosidase and arylsulfatase in soils intending to reduce the cost and time necessary for the realization of these analyses.

Materials and Methods

Soil samples:

30 soil samples collected from different soil types with soil organic carbon varying from 6.5 to 21.5 mg kg⁻¹.

Treatments:

1. Original method (Tabatabai, 1994)
2. Modified method*
3. Modified method* with 50% of original substrate concentration for phosphatase and arylsulfatase; 10% of original for β -glucosidase.

*Modified method:

- (1) reduction in 50% the quantity of soil and reagents;
- (2) use of 15 ml test tubes instead of 50 ml Erlenmeyers;
- (3) use of water bath instead of incubators to sample incubation;
- (4) substitution of the filtering step by centrifugation.



Comparison between the original and modified method

Element of the soil enzyme analysis	Method		Advantage of modified method
	Tabatabai (1994)	Modified	
Incubation media	50 ml Erlenmeyer	10 ml test tubes	Less work space and lab glassware
Quantity of soil and reagents	Original	50% of original	Cost reduction
Incubation	Incubator (37°C/1h)	Water bath (37°C/1h)	Lower time to the reaction media attain the enzyme optimum temperature
Supernatant obtainment	Filtering	Centrifugation	Lower analysis cost and faster procedure

References

Tabatabai, M.A. 1994. Soil Enzymes. p. 775-833. In R.W. Weaver, et al. (ed.) Methods of soil analysis. Part 2. Microbiological and biochemical properties. SSSA, Madison, WI.

Results and Discussion

- All three enzymes showed high correlation between the original and modified methods (Fig. 1).
- The average enzyme activity using the modified method was 30% to 40% higher compared with the original methods. These differences were attributed to the use of water bath for incubation of samples, which results in faster temperature equilibrium of the reaction medium (i.e., more time of incubation under enzyme optimum temperature).
- Reducing the substrate concentration to 25 mM or 10 mM (β -glucosidase) resulted in lower enzymatic activity for all three enzymes, but results were highly correlated with those at 50 mM using the original method.

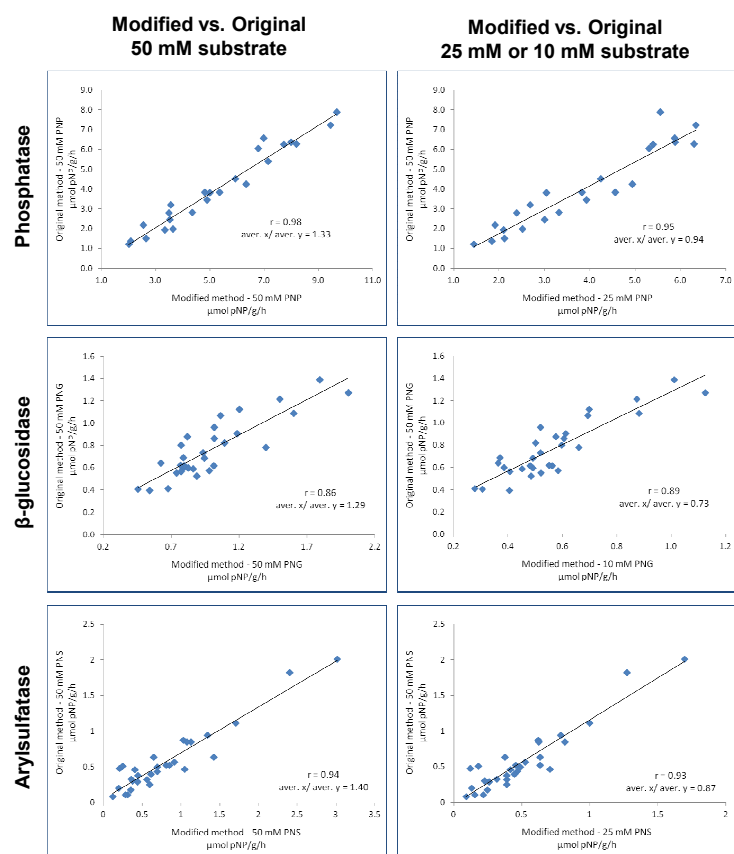


Fig 1. Relationship between phosphatase, β -glucosidase and arylsulfatase activities obtained by the original method and the modified method using either 50 mM or 25-10 mM substrate concentration.

Conclusions

- The modified methods for the analysis of phosphatase, β -glucosidase and arylsulfatase in soils produce results comparable to the original methods described by Tabatabai (1994), and may significantly reduce the costs of these analyses, opening the opportunity of their inclusion in routine soil tests.

Acknowledgements

- This work was partially supported by the Brazilian National Council of Technological and Scientific Development (CNPq, n°485034/2012-3).