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Extraction abilities of methods used for estimating of different phosphorus fractions

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Introduction
There are many fractions of P in soil. All of them can influence the availability of P for plants. Therefore, it is important to have a good knowledge about all P fractions to gain a better insight of processes taking place in soils.

The lowest content of P is usually in soil solution. This fraction is possible to estimate with 0.01M $CaCl_2$ (Houba et al. 1994).

Exchangeable sorbed P (P_{ex}) is another important source of bioavailable P. It is the part of soil P that is exchangeable and sorbed on the surface of soil particles. It can be easily released to the soil solution and become bioavailable. This fraction is possible to estimate via anion exchange (AE) membranes (Kuono et al. 1995, Sato and Comerford 2006).

The most commonly methods used are those for estimating total bioavailable P in soils: e.g., Mehlich 3 (Mehlich 1984), Olsen (Olsen et al. 1954), CAL and Doppel-Lactat (DL) (Hoffman 1991), etc. These extractions are focused on the determination of potentially quickly bioavailable P. It means P in soil solution and weakly bound P.

One of the most important fractions in noncalcareous soils are phosphates sorbed on iron and aluminum compounds (P_{FeAl}). These phosphates are possible to estimate with different fractionation methods, e.g., Chang a Jackson (1957), Hedley et al. (1982) or separately with the Schwertmann (1964) method.

Several fractionation methods, e.g., Chang and Jackson (1957), Marks (1977), Hedley et al. (1982) etc., are commonly used for estimating of total inorganic P (P_{in}). One part of these fractionations is ever estimating of total P (P_{tot}). Based on these data, it is possible to calculate amount of total organic P (P_{org}) as the difference between P_{tot} and P_{in}.

There have been many methods published for estimating total P (P_{tot}) in soils. Some of them are closely described in the work of Kara et al. (1997). Another commonly used extraction procedure is the *aqua regia* extraction (P_{ar}). This method after Crossland et al. (1995) correlates well with alkaline fusion. Therefore, the *aqua regia* extraction is useful for indirect estimation of total P in soils.

The aim of this work was to evaluate selected methods used for estimating of different P forms in soil. The methods studied included 0,01M $CaCl_2$ extracts, anion exchange membranes extracts, Mehlich 3 and Doppel-Lactat methods, estimating of Fe-, Al- sorbed P, fractionation after Marks (1977) and the *aqua regia* extracts.

Material and methods
Soil samples (0-30 cm) were taken from 18 different soil-climatic sites from long-term field experiments in the Czech republic (12 sites) and Germany (6 sites). The pH of the sites ranged between 5.0 and 6.9.

The experiments have been running for more than 20 years at the time of sampling. Commonly were taken 110 samples from different fertilizing treatments. More fertilizing treatments were chosen, because of get higher variability of studied samples. Samples were air-dried and sieved through a 2-mm sieve.

Soil samples were analyzed for different P fractions. The 0.01M $CaCl_2$ extraction (Houba et al. 1994) was used for estimating readily available P (P_{CaCl2}). Anion exchange (AE) membranes adapted after Kuono et al. (1995) were used for the extraction of exchangeable P (P_{ex}). Mehlich 3 (Mehlich, 1984) and Doppel-Lactat (DL) (Hoffman, 1991) methods were used for estimating bioavailable P (P_{M3} a P_{DL}). In order to determine the fraction of P sorbed on Fe and Al compounds (P_{FeAl}), the extraction after Schwertmann (1964) was applied. Fractionation after

Marks et al. (1977) was used for estimating total inorganic (P_{in}), total organic (P_{org}) and total P ($P_{M\text{-}tot}$) contents. *Aqua regia* extracts were used for measuring rezidual P content (P_{ar}). All reagents used were in per analysis quality.

All extract were measured spectrofotometrically using SKALAR SAN[PLUS] SYSTEM.

Results and discussion

The obtained data did not meet normal distribution with the statistical tests. Therefore, basic statistically characteristics (Average, median, maximal and minimal contents) were used for the evaluation of results. Median comparison appeared to be best performance for the evaluating the extraction ability of different methods.

Table 1 shows the percentual distribution of different P fractions in soils based on median comparisons. According to the low contents of $P_{M\text{-}tot}$, P_{ar} was chosen as 100%.

Table 1: Comparison of different extractions methods (P_{ar}=100%)

P_{CaCl2}	P_{ex}	P_{DL}	P_{M3}	P_{in}	P_{org}	$P_{M\text{-}tot}$	P_{FeAl}
0.1%	10%	12%	17%	25%	38%	60%	62%

The lowest percentage of different fractions represented P in soil solution (0.1%). Similar values of 10% and 12%, respectively were reached with AE membranes and the Doppel-Lactat method. The amount estimated with the Mehlich 3 method was 17%. That corresponds well with results of many authors, that the content of bioavailable phosphates in soil is relatively low and reaches usually about 10% of total P content (Marschner 1995, Blume et al. 2002, Bünemann et al. 2007 and others). The P_{org} content was about 13% higher compared to the content of P_{in}. More than 50% of P_{ar} constituted of phosphates sorbed on iron and aluminum compounds. Our results showed that the Marks fractionation did not probably estimate the corresponding P fractions. This is possible to derive from the fact that the P_{FeAl} content was very close to $P_{M\text{-}tot}$. Therefore, more then all P was sorbed to Fe- and Al- compounds, which is not probable. Additionally, the $P_{M\text{-}tot}$ content was significantly lower compared to P_{ar} content. It is clear, that the Marks fractionation is usable only as a rough tool for estimating the ratios between P_{in} and P_{org} in the analyzed soils. A similar ratio between P_{in} and P_{org} was published by Bünemann et al. (2007) using Saunders and Williams extraction method.

Correlation analysis was used for comparison of different P fractions (Table 2). Spearman correlation coefficients, which are suggested for comparison of data with non-normal distribution were used for the calculating of relations. The relations were calculated at p-values of 0.01 and 0.001.

Table 2: Spearman correlation coefficients for different methods

	P_{CaCl2}	P_{ex}	P_{DL}	P_{M3}	P_{in}	P_{org}	P_{M-tot}	P_{FeAl}	P_{ar}
P_{CaCl2}	1.00	0.50**	0.59**	0.67**	-0.06	-0.39*	-0.24	-0.08	-0.10
P_{ex}		1.00	0.82**	0.45**	0.23	0.08	0.18	0.26*	0.26*
P_{DL}			1.00	0.58**	0.33*	0.06	0.22	0.36*	0.36*
P_{M3}				1.00	0.46	-0.14	0.19	0.43**	0.32*
P_{in}					1.00	0.61**	0.91**	0.96**	0.91**
P_{org}						1.00	0.89**	0.66**	0.66**
P_{M-tot}							1.00	0.91**	0.88**
P_{FeAl}								1.00	0.94**
P_{ar}									1.00

** significant relations at p-value 0.001
* significant relations at p-value 0.01

Comparing the different methods for estimating bioavailable P fractions in soils (P_{CaCl2}, P_{ex}, P_{DL} a P_{M3}) led to close correlations at p-value 0.001. It means that with the mentioned methods it is possible to estimate mutually dependent P fractions and in this case it shows close relations between P in soil solution and exchangeable and total bioavailable P, respectively.

In most of cases, statistically significant relations between bioavailable forms of P and P_{org} were not reached.

High correlation coefficients between P_{in}, P_{org} and P_{M-tot} were obtained, because these fractions originated from one fractionation. The correlation coefficient between P_{org} and P_{M-tot} was even 0.89.

Close relations between P_{FeAl} and P_{M-tot} and P_{ar} showed that a considerable content of P in noncalcareous soils can be sorbed on Fe and Al compounds.

Aknowledgements

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