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COMPARISON OF SUITABLE EXTRACTANTS USED IN SOIL PHOSPHORUS TESTING ON DIFFERENT SOILS

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ABSTRACT

The present study was carried out to compare the six Phosphate extractants applied on fourteen different soil types. The extractants used include Acetic acid, Sodium acetate buffer (Morgan solution), Ammonium acetate buffer (Modified Morgan's solution), Sodium bicarbonate (Olsen extractant), Acidified ammonium fluoride (Bray – I reagent) and Anion Exchange Resin (AER). The fourteen soil were different in soil properties as pH, Organic matter content, Fe, Al and soil texture. The soil were grouped in to two on the basis of pH (acidic and alkaline). The results showed that Phosphate extracted by various extractant showed significant difference at 1% level of probability. The highest Phosphate concentration was measured in different soil by using the Bray I extractant. The lowest was determined in AER. Significant correlations have been found between the extractant except for Bray which showed non – significant correlation. Comparing the effectiveness of soil pH showed positive correlation between the extractants at pH range of 5.2 to 6.3. While at higher pH in range of 6.3 to 7.9 was non – significant. It may be concluded from the result that Bray I was effective in extracting P concentration from soil in acidic condition while Acetic acid and Sodium Bicarbonate (Olse reagent) were more effective under slightly acidic to alkaline condition.

Keywords: Soil, Phosphate, Extractants, Alkaline soils, Acidic soils, correlations.

INTRODUCTION

Phosphorus (P) is an important plant nutrient required in relatively large amount to perform various functions of plant. Due to increase in the cost of phosphatic fertilizers and the desire for environmental correct usage of fertilizers, there is a renewed interest in developing sound system for testing soils to make fertilizer recommendations. Methods for determining soil phosphorus and its availability to plants are essential for developing principles and knowledge of the nature and behavior of P in soil. The purpose of these methods has been to characterize the P in soil system. Soil testing procedures are being used all over the world with varying degree of success to determine the P status and assist in making fertilizer recommendations. A major limitation is that the results are influenced by the soil type with the result that extensive field calibration is often required. There are several soil characteristics which may affect the Phosphate availability to plants as well as the Phosphate extraction by different chemicals. These characteristics includes extractable Al, Fe, soil clay content, lime, organic matter, soil pH etc. Haney *et al*. (2006) reported that there is great variation in soil therfore developing a single method of extraction may be an expensive and time conuming, hence identifying an extractant that has more accuracy with respect to plant available nutrient is required.Variation amongst the P content in grassed and non – grassed soil by using the different extractants have been reported by Khan *et al*. (2013).

In the tropical soils extractable Fe and Al oxides are important (Agbenin, 2003). Although, each of the extracting method used has problem in different soils, chemical extractants are used according to the soil characteristics, this determine the effectiveness of the extractants, otherwise it may lead to test results difficult to be interpreted (Myers *et al*., 2005). The present study focus on the comparison of soil phosphate extractants for different soil and their correlation with each other and soil properties.

MATERIALS AND METHODS

Soil samples were collected at random from various sites of Scotland, United Kingdom (UK). The soil were attributed to soil series using the soil Memoirs and soil maps for each area. Where the areas were not mapped, Soil Memoirs for an adjacent area were used (Mitchell and Jarvis, 1956; Gran *et al*., 1962; Ragg *et al*., 1976). The Physico – chemical characteristics of soil including pH (McLean, 1982), % LOI (Davies, 1974) and % O.C (Nelson and Sommer, 1996) were determined.

Also the extractable Fe and Al was extracted with acid ammonium oxalate (Tamm's reagent) (Klute, 1996) and Soil texture was determined using the hydrmetric method (Bouyoucos, 1962). The physico – chemical characteristics of the soil are described in table 1.

Extraction Methods for soil Phosphate: Six different procedures were selected for extraction of soil phosphate. These procedures are widely used for estimation of available soil phosphate, in the domain of agriculture. All the procedures for getting soil extracts, were applied to soil samples in the same manner, without modification from the original. All the soils were extracted in triplicate with each extractants. The extractants used in the study were 0.5 M Acetic acid (Korndorfer *et al*., 1995), Sodium acetate buffer (Morgan, 1941), Ammonium acetate buffer (McIntosh, 1969), Sodium bicarbonate (Olsen *et al*., 1954), Acidified ammonium fluoride (Bray and Kurtz, 1945) and Anion Exchange Resin (Sibbesen, 1978).

Table 1. Physico – chemical characteristics of soil.

S. No.	Soil	Soil Characteristics					
		pH	LOI %	O.C. %	Fe %	Al %	Texture
1	DreghornNo. 1A	5.2	4.8	1.84	0.29	0.33	Sandy Loam
2	DreghornNo. 1B	5.5	2.2	0.68	0.25	0.36	Loamy sand
3	Darvel A	5.7	8.8	2.95	0.85	0.42	Sandy Clay Loam
4	Caprington A	5.9	8.4	3.41	0.52	0.17	Sandy Clay Loam
5	Bargour A	5.5	5.8	1.87	0.63	0.35	Sandy Loam
6	Dunlop A	4.9	18.4	6.69	1.08	0.58	Clay Loam
7	Peat (Blackstoun)	5.4	20.8	8.41	1.11	0.45	Clay Loam
8	Dreghorn No. 2A	5.8	8.8	3.51	0.96	0.43	Sandy Loam
9	Dreghorn No. 2B	6.1	5.0	1.20	0.67	0.35	Sandy Loam
10	Midelney (Grass) A	6.3	15.6	6.18	0.74	0.11	Clay
11	Midelney (Arable) A	7.2	11.4	4.19	0.59	0.11	Silty Clay
12	Midelney (Arable) B	7.6	5.8	1.67	0.33	0.11	Silty Clay
13	Alluvium A	7.4	6.2	2.78	0.41	0.08	Sandy silt loam
14	Swaffham Prior A	7.9	3.2	1.28	0.11	0.10	Sandy Loam

LOI = Loss on ignition, O.C. = Organic carbon

RESULTS AND DISCUSSİON

Estimation of soil phosphate by extractants: The mean amount of PO_4 – P extracted from the soil by six extractants are given in table 2. The amount of phosphate extracted showed a wide variation between soils. The statistical analysis of the data showed that the amount of PO_4 – P extracted by various extractants varied significantly. The highest PO_4 – P was removed by Bray I reagent, followed by acetic acid, which indicate that these methods removed some of the unavailable P, Gikonyo *et al*. (2010) reported Bray I and Mehlic 3 extractants removed that highest Phosphorus from both the fertilized and non fertilized and considered as these are acidic extractants and may remove the water insoluble P sorbed by the soil.

The performance of sodium bicarbonate, sodium acetate and ammonium acetate is comparable. This suggests that three methods probably extract some of the same kind of soil P. The lowest amount of PO_4 – P was recorded in anion exchange resin, which is analogous to plant root. However, Anion Exchange resin is considered to be a good method for the plant available P and it has the capability to extract P irrespective of soil properties. Similar finding have been reported by Raven and Hossner, 1993.

Correlation between extractants for estimation of extractable soil phosphate: Correlation coefficient between six extractants, for all soil under study. It was observed that Bray reagent showed non – significant correlation with all the other extractants under study (Table 3). The relationship of Bray I test was affected by soil characteristics, particularly by oxalate extractable Al. The Bray I procedure performed erratically in calcareous soils as compared to others in the study.

Similar results were obtained by Van Lierop and Tran (1990). The three extractants: ammonium acetate buffer, sodium acetate buffer and 0.5M sodium bicarbonate revealed best relationship, in all the 14 soils. Wuenscher *et al*. (2015) reported that most Phophorus extractants showed good correlation in their study of central Europe soils. This suggest that these extractants have similarity in selectivity of dissolution of same kind of phosphatases (Chang and Juo, 1963). Anion exchange resin showed better correlation with sodium bicarbonate, good correlation with ammonium acetate buffer as well as with sodium acetate buffer and non significant correlation with other two extractants.

Sodium bicarbonate also behaved like anion exchange resin, in having no significant correlation with acetic acid and acidified ammonium fluoride (Bray I

reagent). However, it indicated better correlation with ammonium acetate and anion exchange resin.

Comparing the effectiveness of extractants at different pH: Keeping in view the general trend of soils in the extractable phosphate relationship, the soil were split into two groups, on the basis of pH, 1st range 5.2 – 6.3 and 2nd range 6.3 – 7.9 (Table 4 and 5). It was observed that correlation between extractants in soil having pH below 6.3 were improved. But the correlation between extractants were decreased in alkaline soils. Zbiral (2000) reported a strong influence of soil pH on the acidic extraction methods.

Table 2- Soil phosphate content by using different extractants.

S. No.	Soil	Extractants (µg g^{-1} soil)					
		NH_4 – acetate	Na – Acetate	Acetic acid	Na – Bicarbonate	Bray I	AER
1	DreghornNo. 1A	5.3	6.2	8.9	15.9	68.5	7.1
2	DreghornNo. 1B	0.5	1.4	1.5	4.2	31.7	1.4
3	Darvel A	11.2	13.6	11.5	32.3	114	11.9
4	Caprington A	15.8	14.3	27.2	28.2	64.5	19.9
5	Bargour A	22.6	25.6	29.1	57.2	211	28.5
6	Dunlop A	5	8.3	11.9	26.4	127	4.5
7	Peat (Blackstoun)	8.8	12.7	21.4	38.4	140	8
8	Dreghorn No. 2A	9.5	10.4	23.7	25.2	153	6.4
9	Dreghorn No. 2B	0.7	1.6	0.8	5.2	17.8	0.3
10	Midelney (Grass) A	51.6	44.3	112.0	34.9	93	28.3
11	Midelney (Arable) A	32.4	29.1	69.0	27.0	56.9	13.8
12	Midelney (Arable) B	11.6	3.5	49.7	2.0	2.7	1.9
13	Alluvium A	112.0	113.0	113.0	81.0	0.8	20.1
14	Swaffham Prior A	80	68.5	133.0	37.9	0.6	12.5
	Means	26.3 c	25.2 c	43.8 b	29.7 c	77.2 a	11.8 d

Means followed by similar letter(s) do not different significantly at 1% level of probability.

Table 3- Correlation coefficient between extractable soil phosphate using six extractants from 14 soils.

Extractants	NH4 – acetate	Na – Acetate	Acetic acid	Na – Bicarbonate	Bray I
Na – Acetate	0.99***				
Acetic acid	0.90***	0.85***			
Na – Bicarbonate	0.75**	0.81***	0.53NS		
Bray I	- 0.38NS	- 0.32NS	−0.34NS	0.23NS	
AER	0.54*	0.55*	0.53NS	0.73**	0.32NS

NS Non – significant at 5% * Significance at 5% **Significant at 1% *** Significant at 0.01%

Table 4 - Correlation coefficient between soil phosphate using six extractants from 9 soils having pH 5.2 to 6.3.

Extractants	NH4 – acetate	Na – Acetate	Acetic acid	Na – Bicarbonate	Bray – I
Na – Acetate	0.97***				
Acetic acid	0.88**	0.86**			
Na – Bicarbonate	0.89**	0.97***	0.83**		
Bray – I	0.73*	0.83**	0.75*	0.90***	
AER	0.97***	0.93***	0.80*	0.83**	0.61NS

NS Non – significant at 5% * Significance at 5% **Significant at 1% *** Significant at 0.01%

Table 5- Correlation coefficient between soil phosphate using six extractants from 5 soils having pH 6.3 to 7.9.

Extractants	NH4 – acetate	Na – Acetate	Acetic acid	Na – Bicarbonate	Bray – I
Na – Acetate	0.99***				
Acetic acid	0.82NS	0.76NS			
Na – Bicarbonate	0.95*	0.98*	0.67NS		
Bray – I	-0.31NS	-0.29NS	-0.01NS	-0.16NS	
AER	0.52NS	0.54NS	0.62NS	0.621NS	0.636 NS

NS Non – significant at 5% * Significance at 5% **Significant at 1% *** Significant at 0.01%

Acid ammonium fluoride extractant (Bray) yielded the highest Phosphate extracted in this study, but it removed negligible amount of phosphate from alkaline soil. The relationship between pH and extractable Bray - I P was negatively significant at 1% level of probability ($r=-0.67^{**}$) in all 14 soils. In the relationship studies of various soil characteristics with extractable Bray – I P, it revealed significant correlation of extractable P with pH, Al and Fe. But no significant correlation with LOI, organic matter and soil texture. Morgan *et al*. (2010) found that Bray I extractants yielded P concentration similar to water and carbonate extractable soil P concentration.

Acetic acid ranked second in extracting highest amount of phosphate from soils. Contrary to Bray it removed higher amount of PO_4 – P from alkaline soils, presumably due to larger proportion of calcium phosphate (Fig. 1). It showed a very highly significant correlation with ammonium acetate buffer ($r = 0.90^{***}$) and sodium acetate buffer ($r=0.85^{***}$). This methods is widely used as index of available phosphate supply in soils throughout the world. Acetic acid extractable PO_4 – P has very highly significant correlation with pH($r=0.78^{***}$) and Fe concentration (r=-0.78), while correlation with other soil characteristics were found non – significant.

Sodium bicarbonate (Olsen *et al*., 1954) was developed for calcareous soils, but in this study its performance was found fairly good for acidic soil as well as alkaline soils. According to Olsen *et al*. (1954) there are two major mechanisms in $NaHCO_3$ extraction process. Calcium phosphates increase in solubility in $NaHCO_3$ as a result of the repression of Ca activity and HCO_3^- and CO_3 and OH^- ions replaces P ions on the surface soil particles. Non – significant relationship of NaHCO3 extractable P with any of the soil characteristics in both acidic and alkaline soil. Morgan *et al*. (2010) reported that Olsen P underestimated that the P concentration in acid soil. Wijebandara 2004 reported poor correlation of $R^2= 0.460$ ($P<0.05$) in acid soils. Olsen extractants generally gave higher value of P concentration in soil having greater pH (Ottobong *et al*., 2004 and Eriksson, 2009). But Contrary to these findings Frank *et al* 1998 found that Olsen P gave rational results for soil without Calcium carbonate.

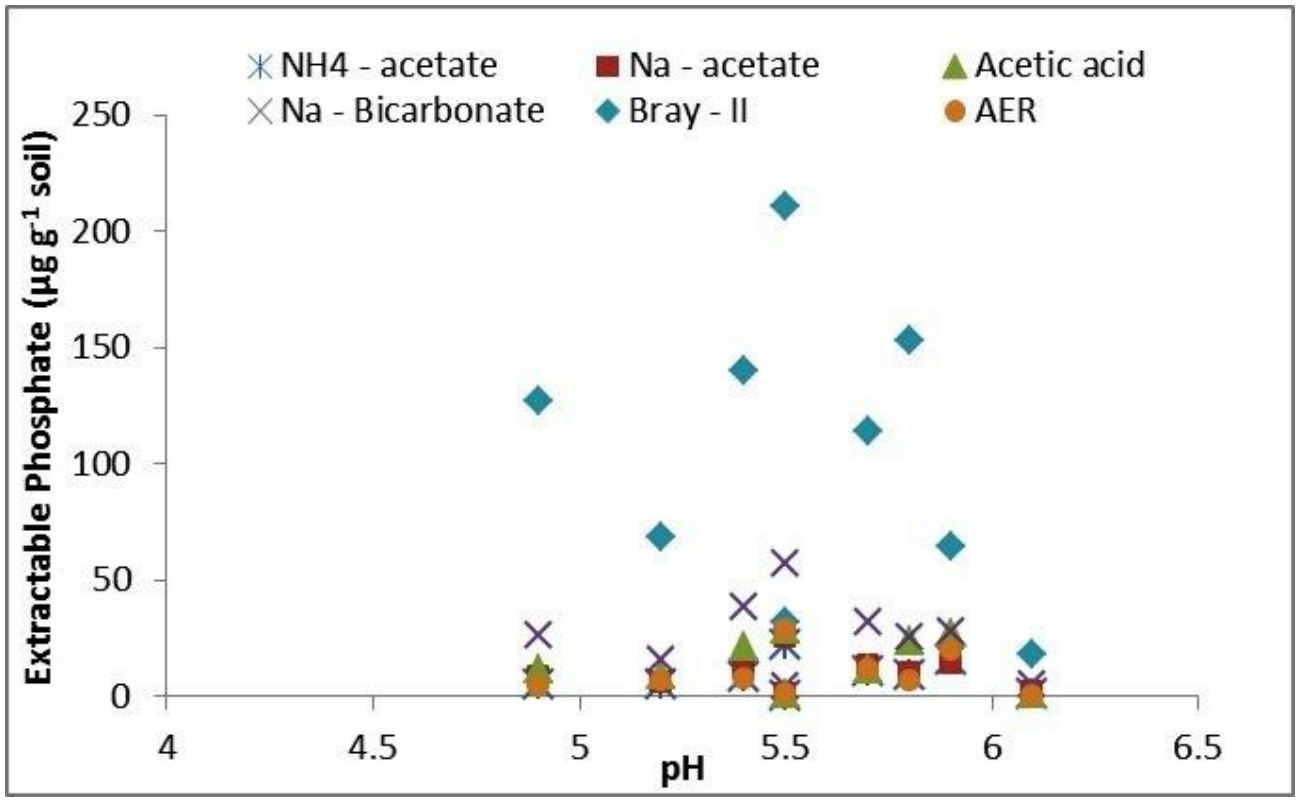

Fig.1: Extractable Phosphate extracted by different methods of soils having 5.2 to 6.3.

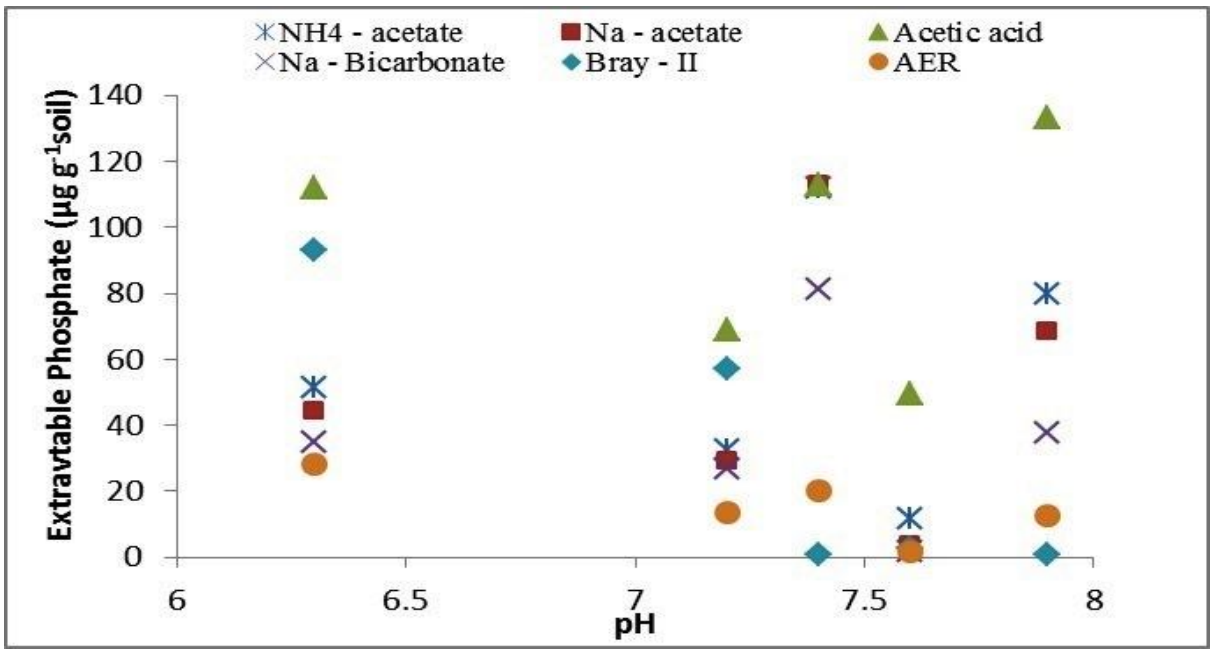

Fig. 2: Extractable Phosphate extracted by different methods of soil having pH 6.3 to 7.9.

The amount of PO_4 – P extracted by sodium acetate buffer (Morgan's reagent) was statistically comparable with sodium bicarbonate and ammonium acetate buffer. This method show very highly significant correlation with ammonium acetate ($r = 0.99^{***}$), 0.5 M acetic acid ($r= 0.85^{***}$), sodium bicarbonate ($r= 0.81^{***}$) and significant with anion exchange resin ($r = 0.55^{*}$) in all 14 soils under study. The amount of PO_4 – extracted by this method has no – significant correlation with the soil characteristics except pH and Fe which has significant coefficient of correlation of 0.61^{*} and -0.59^{*} respectively. The amount of Phosphate extracted by Morgans method was lower than that amount extracted by Bray and Olsen reagent in acidic soil, however it was relatively higher in alkaline soil.

Ammonium acetate buffer (pH 4.8) performed almost similar to the Morgan's method (Fig. 2). The PO_4 – P extracted by this method did non significantly differ from ammonium acetate and sodium bicarbonate at 1% level of probability. The Coefficient of correlation of this method with sodium acetate, acetic acid, $NaHCO_3$ and anion exchange resin found 0.99^{***}, 0.90^{***}, 0.75^{**} and 0.54^{*} respectively. The relationship of extractable PO_4 – P with soil properties (pH and Fe) was observed highly significant. In alkaline soil the Fe was found negatively correlated.

Anion exchange resin extracted the lowest quantity of phosphorus. By comparing the Anion exchange resin with the other extractants showed that it has good correlation with ammonium acetate ($r=0.55^{*}$) and sodium acetate ($r = 0.55^{*}$), better correlation with $NaHCO_3$ ($r= 0.73^{**}$) and non – significant correlation with acetic acid and Bray for all 14 soils. Anion exchange resin extractable P did not show any significant relationship with all the soil characteristics under study.

Conclusion: It may be concluded from the research that comparing the different phsphate extractants on different soil showed the highest P concentration in various soil by the Bray I followed by Olsen extractant. The lowest P concentration was recorded in AER. Correlations between the extractant was significant amongst all the extractants except for Bray I which was non – significant. It was also found from the results of the experiment that at low pH 5.2 to 6.3 Bray I extractant was more suitable followed by Olsen extractant. While at pH range 6.3 to 7.9 Acetic acid and Na – Bicarbonate (Olsen reagent) were more effective and have extracted greater P as compared with the rest of the extractant. The least value at higher pH was observed in Bray I.

REFERENCES

Agbenin, J.O. and B. V. Raij (2001). Kinetics and Energetics of phosphate release from tropical soils determined by mixed ion-exchange resin. Soil Sci. Soc. America. 65:1108-1114.

Bouyoucos, G.J. (1936). Directions for Making Mechanical Analysis of Soils by the Hydrometer Method.Soil Sci. 4:225 – 228.

Bray, R.H. and L. T. Kurtz (1945). Determination of total, organic, and available forms of phosphorus in soils. Soil Sci. 59:39 – 45.

Chang, S.C. and S. R. Juo (1963). Available phosphorus in relation to forms of phosphatase in soils. Soil Sci. 95: 91 – 96.

Davies, B. E. (1974). Proc. Soil Sci. Soc. America., 38: 150 - 151.

Eriksson, A. K. (2009). Phosphorus in agricultural soils around the Baltic Sea. M.Sc. Thesis. Swedish University of Agricultural Sciences. Sweden.

Frank, K., D. Beegle and J. Denning (1998). Phosphorus. In: Brown J.R. (ed.): Recommended Chemical Soil Test Procedures for the North Central Region. Columbia, North Central Regional Research Publication 221, Missouri Agricultural Experiment Station, 21–30.

Haney, R.L., E.B. Haney, L.R. Hossner and J.G. Arnold (2006). Development of a new soil extractant for simultaneous phosphorus, ammonium and nitrate analysis. Commun. Soil Sci. Plant Anal. 37:1511-1523.

Khan, Q. U., T. H. Flower and Sanaullah (2013). Influence of soıl phosphorus on enzyme actıvıty, ryegrass yıeld and extractable phosphate. The J. Animal and Plant Sci. 23(1): 222 – 226.

Klute, A. (1996). Methods of soil analysis. Madison. American Society of Agronomy. Agronomy Monograph 9.

Korndorfer, G.H., D. L. Anderson, K. M. Portier and E. A. Hanlon (1995). Phosphorus soil test correlation to sugarcane grown on Histosols in the Everglades. Soil Sci. Soc. America J. 59:1655 – 1661.

Kowalenko, C.G. (2010). Relationships between Extraction Methods for Soil Nutrient Testing in British Columbia. Agriculture and Agri-Food Canada. pp. 25.

Grant. R., C. J. Bown and E. L. Birse (1962). Map sheet 14, Ayr. Soil Survey of Scotland. The Macualay Institute for soil research, Aberdeen.

Mclean, E.O. (1982). Soil pH and lime requirement. In: Miller, A.L.R.H. and D.R. Keeney (eds.). Method of soil Analysis part 2. American Society of Agronomy. No. 9. 199- 209. Madison, WI, USA.

McIntosh, J. L. (1969). Bray and Morgan soil test extractants modified for testing acid soils from different parent materials. Agron. J. 61:259-265.

Mitchelle, B. D. and R.A. Jarvis (1956). The soil of the country round Kilmarnock. Memoirs of the soil survey of Great Britain Soil Survey of Scotland. The Macualay Institute for Soil Research, Aberdeen.

Morgan, M.F. (1941). Chemical soil diagnosis by the universal soil testing system. Conn. Agric. Exp. St. Bull. No. 450.

Morgan, K.T., S. Sato, K. Mahmoud and A. Zekri (2010). Evaluation of soil extractants on sandy soils with high calcium concentrations in southern Florida.19thWorld Congress of Soil Science, Soil Solutions for a Changing World, 1 – 6 August 2010, Brisbane, Australia.

Myers, R.G., A. N. Sharpley, S. J. Thien and G. M. Pierzynski (2005). Ion-Sink phosphorus extraction methods applied on 24 soils from the continental USA. Soil Sci. Soc. America J. 69: 511- 521.

Nelson, D. W. and L. E. Sommers (1996). Total Carbon, Organic Carbon, and Organic Matter. In: Methods of Soil Analysis, Part 3. Chemical Methods,

Olsen S.R., C. V. Cole, F. S. Watanabe and L. A. Dean (1954). Estimation of available Phosphorous in Soil by extraction with Sodium Bicarbonate, USDA Circular. 939.

Otabbong E., P. Leinweber, A. Schlichting, R. Meissner, M. Shenker, I. Litaor, A. Sapek, S. Robinson, A. Nidermeier, H. Hacin and J. R. Otabbong (2004). Comparison of Ammonium Lactate, Sodium Bicarbonate and Double Calcium Lactate Methods for Extraction of Phosphorus from Wetland Peat Soils. Acta Agric. Scan. 54(1): 9-13.

Rag, J. M., B. M. Shipley, N. A. Duncan, J. S. Bibby and D. W. Merrilees (1976). Map sheet 31, Airdris. Soil Survey of Scotland. The Macualay Institute for Research, Aberdeen.

Sibbesen, E. (1978). An investigation of the anion exchange resin method for soils phosphate extraction. Plant Soil. 50: 305-321

Van Lierop, W. and T. S. Tran (1990). Relationship between crop response and available phosphorus by Kalowna EDTA and DTPA modified multiple element extractants. Soil Sci. 149: 331 – 338.

Wijebandara, D.M.D.I. (2004). Soil Phosphorus test methods calibration by field experimental data for the soils of low country intermediate zone of Sri Lanka. COCOS 16: 65 –73.

William, E.G. and A. H.Knight (1963). Evaluation of soil phosphate status by pot experiments, conventional extraction methods, labile phosphate values estimated with aid of phosphorus – 32. J. Sci. Food Agric. 14:555 – 563.

Wuenscher, R., H. Unterfrauner, R. Peticzka and F. Zehetner (2015). A comparison of 14 soil phosphorus extraction methods applied to 50 agricultural soils from Central Europe. Plant Soil Environ. 61(2): 86 – 96.