ASSESSMENT OF SOIL PHOSPHORUS TESTS FOR SITUATIONS IN AUSTRALIA WHERE REACTIVE PHOSPHATE ROCK AND WATER-SOLUBLE FERTILIZERS ARE USED

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Abstract. A selection of commonly used soil P tests, which included anion and cation exchange resin membranes, were compared in a glasshouse experiment using subterranean clover, and evaluated in the field at 19 sites from the National Reactive Phosphate Rock Project in 1993 and at 6 sites in 1995. The ability of the soil P tests to predict plant response was used to evaluate the tests. In the glasshouse experiment, the resin test was less effective than the Bray and Colwell tests in its ability to assess the level of plant available P from the different fertilizer treatments. Seventy one percent of the variation in total P content of the subterranean clover tops was explained by resin extractable P values, whereas the Colwell procedure accounted for 81% and the Bray 1 procedure accounted for 78%. Water and $CaCl_2$ extracts were poor predictors of P content. In the field experiments, all tests evaluated performed poorly in describing the relationship between soil test P and the level of P applied and relative yield and soil test P over a wide range of soil types and environments. The best performing test was the Bray 1 test, though the relationship was poor.

1. INTRODUCTION

The existence of first year only applications of fertilizer in one of the core experiments [1] established at each site in the National RPR Project provided the opportunity to compare different soil P testing procedures. This was done by relating soil P test values in soil samples collected at the beginning of the growing season to the pasture yields measured in that growing season. The treatments included large applications of triple superphosphate, North Carolina phosphate rock (NCPR), and a partially acidulated NCPR.

The soil testing procedures used were the Bray 1, Colwell, water, $CaCl_2$ and mixed CER/AER exchange resins. The comparisons were made in a glasshouse pot experiment undertaken in 1993 using subterranean clover and a soil collected from one of the field sites, and 19 field experiments in 1993 and 6 field experiments in 1995.

2. MATERIALS AND METHODS

2.1. Soil P tests

The soil P tests evaluated were anion and cation exchange membrane (AER-CER) [2], 0.01 M $CaCl_2$, water, Colwell [4], and Bray1 [5]. For AER-CER, a soil solution ratio of 1:40 was used with a 16 hr shaking time. The cation and anion exchange resin membranes (BDH products) were prepared using 1 mol/L NH_4Cl. Phosphate ions were eluted from the AER with 40 mL of a 0.25 mol/L $BaCl_2$ and 0.1 mol/L HCl solution using a 0.5 hr shaking time. Phosphorus in the eluent was determined with a spectrophotomer using a $SnCl_2$ reduced ammonium molybdate — HCl solution. The 0.01M $CaCl_2$ and water methods used a soil solution ratio of 1:10 with a 16 hr shaking time. The suspension was centrifuged for 15 min. at 7000 rpm and the supernatant was passed through a Whatman No. 42 filter paper. The first few drops of the filtrate were discarded. The concentration of P in the supernatant was determined with a spectrophotometer using the Malachite green method [3].

This paper has been modified from the original article "Assessment of soil phosphorus tests for situations in Australia where reactive phosphate rock and water soluble fertilizers are used" by Simpson, P.G., Weatherley, A.J., Sale, P.W.G., McLaughlin, M.J., Hoy, V. and Lancaster, P. published in in Aust. J. Expt. Agric. 37 (1998) 1027-1035 by permission of CSIRO Publishing.

For all of the soil P tests the <2 mm fraction of the top 10 cm of soil (pot and field experiments) was used. Soil samples from the pot trial were taken from the full depth of the pot. In the field experiments, soil samples (0-10 cm) were collected by bulking 6 to 10 2-cm soil core samples taken from the central portion of each plot before the start of the growing season in 1993 and 1995. The start of the growing season occurred in the autumn in southern Australia, and the spring in northern Australia.

2.2. Glasshouse pot trial

2.2.1. Treatments and experimental design

Triple superphosphate (TSP), North Carolina PR (NCPR), and a partially acidulated PR (PAPR) made by 50% acidulation of North Carolina PR with sulfuric acid, were applied at levels to produce a range of soil P levels. Some properties of the fertilizers are provided by Simpson *et al.* elsewhere [1]. A soil with pH of 4.0 (0.01 mol/L $CaCl_2$) and a Colwell P of 29 µg/g collected from Upper Ryans Creek, near Benalla, Victoria, was used for the experiment. Soil, taken from the top 10 cm of an established pasture, was sieved (<2 mm) and weighed out into 6.4 kg lots and placed into plastic bags. After the addition of basal nutrients (equivalent kg/ha: 5 of N, 40 of K, 10 of Mg, 2 of Zn, 1 of Cu, 0,1 of Mo, and 1 of B), the P fertilizers were added at equivalent levels of 10, 20, 30, 40, 50, 60, 70, and 80 kg/ha of P for the TSP and PAPR and 10, 20, 30, 40, 50, 60, 80, and 160 kg/ha of P for the NCPR. A nil P treatment was included as a control. All fertilizers were ground to pass a 0.25-mm sieve to achieve an even distribution of fertilizer through the soil. The fertilizers and soil were mixed thoroughly in the plastic bags and placed in 30-cm diameter pots in the plastic bags and so were not free draining. After wetting to 90% field capacity, the soils were allowed to incubate at glasshouse temperature (25°C) for 8 weeks. Pots were then sown with inoculated subterranean clover (*Trifolium subterranean* cv. Trikkala) seeds, with plants later being thinned to 12 per pot. Pots were arranged in a randomized complete block design, were regularly watered to weight and re-randomized at each watering.

2.2.2. Measurements and data analysis

After incubation, a soil sample was collected to measure soil test P using the procedures described above. Soil test P was then related to level of P application. Three harvests were taken of plant tops up to seed set. Plant tops were dried, weighed and analyzed for P concentration to determine the P content (P concentration multiplied by yield).

Linear regression was used to examine (i) the relationship between level of applied P and soil test and (ii) soil test P and P content.

2.2.3. Field evaluation of soil tests

Soil tests were undertaken in the field using soil samples and pasture dry matter yields from sites included in the National Reactive Phosphate Rock project [1]. This project was established to evaluate the agronomic effectiveness of reactive phosphate rocks in a range of Australian environment [6]. Site locations and information, soil physical and chemical descriptions, and comprehensive details of the experimental design and layout are given in Simpson *et al.* [1]. In summary, the sites were located in the high-rainfall permanent pasture zones across Australia. Soils were acidic (pH < 6.5 water), and representative of major soil types in each region. Soils ranged in texture and included, clay loams, loams, sandy clay loams, sandy loams and acid peaty sands. A comprehensive description of soil chemical and physical properties and site characteristics are presented in Simpson *et al.* [1]. Site identification in the National RPR project was made using the first letter in the name of the state in which they were located, and a number representing an arbitrary position in the series of 30 experiments. e.g. Q2 was a site in Queensland and was the second in the series of 30 sites.

2.2.4. Treatments

Treatments from the National RPR Project used for the field evaluation were those receiving applications of TSP, North Carolina PR, and PAPR applied in the first year only of the trials (1992), and the control (nil P) treatment. Fertilizers were applied at 8X and 16X levels determined for each sites, where the 4X P level was expected to produce 90% of the maximum yield response to TSP fertilizer under optimum conditions. For a more detailed description of how the 'X' level was determined see Simpson *et al.* [1]. The level of application depended upon the environment in which the site was located, and differences between environments meant that there was a wide range of P application levels ranging from 23.2 to 160 kg P/ha. Fertilizers for all treatments were applied in 'as received' form to small field plots (3 m x 5 m or 2 m x 2 m in Tasmania) at the start of the growing season in 1992 only (autumn in southern Australia, and spring in tropical northern Australia). Basal nutrients including sulfur were also applied to each plot [1]. Treatments were laid out in a randomized complete block design. One exception was the sites located in Victoria, where an unbalanced incomplete block design was used. Treatments were replicated 3 times [1].

2.2.5. Measurements and data analysis

Soil samples were collected and processed as previously described. In 1993, soil samples from the control treatments were not analyzed.

Pasture dry matter yields were determined in each growing season by regular harvesting of plots with mowers or rising plate meters [1]. The dry matter yield for each plot was expressed as a % of the maximum (relative) yield at each site. The treatment with the highest mean yield for the experiment, which included the year-1 only treatment, was used as the maximum yield.

Linear regression analysis was used to examine the relationship between soil test P and level of P applied. The relationship between relative yield and soil test P was fitted to a Mitscherlich equation. The goodness of the fit of the non-linear relationship between relative yield and soil test P was assessed from the magnitude of the square root of the residual mean square (RMSE). For reasons presented by Kvålseth [7] and Ratkowsky [8], R^2 was not used as it is considered unsuitable for use in multiple or simple non-linear regression. The smaller the RMSE, the better the agreement was between model and data. In this paper, where relative yield (%) is used, a RMSE of 5% or less was considered to be acceptable variation or error in the fitted Mitscherlich model [9].

3. RESULTS

3.1. Glasshouse experiment

There was a good linear relationship between resin, Colwell and Bray 1 soil test values and the level of P applied (Fig. 1). The Colwell and resin procedures extracted more P from the fertilized soil than the Bray 1 procedure, which may simply reflect the increased extraction time. The Colwell soil test showed some differentiation between fertilizer sources, (Fig. 1b) with the regression line for TSP having a significantly greater slope ($P < 0.05$) than the common line fitted to NCPR and PAPR. There was only a fair correlation between water extractable P and the level of P applied ($R^2 = 0.59$), while the relationship between $CaCl_2$ extractable P and the level of P applied was poor ($R^2 = 0.21$) (data not presented).

Both the water and $CaCl_2$ procedures were poor in describing the relationship between extractable P and plant response (P content), and are not considered further. There were no significant dry matter yield responses to fertilizer P application ($P > 0.05$). However, P content responses to applied P were significant ($P > 0.05$) so P content was used. P content increased linearly with level of fertilizer P applied (Fig. 2). The relationship between P content and the level of P applied was similar for all 3 fertilizers (Fig. 2).

Good relationships were found between P content and soil test P (Fig. 3). The Colwell procedure accounted for 81% of the variation in P content, the Bray 1 accounted for 79%, and the AER/CER resin accounted for 71%.

3.2. Field experiment - 1993

For all sites and fertilizers, all tests were poor in describing the linear relationship between soil test P and level of P applied P ($R^2 < 0.11$) (data not presented). Only when each site was considered separately did any of the tests give a good account of the relationship. Values for R^2 for the separate sites were all greater than 0.6 and ranged from 0.64 to 0.98 (n = 9). Resin, Colwell and Bray 1 performed equally well at most sites (data not presented).

A multiple linear regression was used to examine the variation in soil test P, across all sites for each soil test, as a function of site, level of P applied, and fertilizer type effects. There was a major site effect ($P < 0.05$) where differences in sites accounted for a significant proportion of the variation in soil test P, and lines of best fit for sites would have significantly different intercepts. This was not surprising given the range (between 3 and 46 µg/g 0-10 cm Colwell, [1]) in pre-fertilizer extractable P concentrations measured in soil samples collected from each site before the experiment began. All tests, except the resin test, showed a fertilizer type effect, indicating that the resin test was the only one that did not discriminate between the fertilizer types. Dry matter results from the field trial for 1993 indicated that there was generally no statistically significant difference in dry matter production between the TSP, PAPR, and RPR treatments used (data not presented). These findings are therefore consistent with the resin test result, which indicated that there was no difference between fertilizers.

None of the soil P tests showed a close relationship between relative yield and soil test P for all sites (Fig. 4). The root mean square error (RMSE) value was lowest for the Bray 1 test (16.5%), followed by the Colwell (16.8%), and resin (18.0%) (Table I). The units of RMSE are the same as relative dry matter yield (ie. %) and represent the average variation observed from predicted values. All the RMSE values were larger than that considered acceptable (5%) for non-linear regression.

There was no improvement in the relationship between relative yield and soil test P when each fertilizer was considered separately (Table I). The best relationship occurred for RPR, with RMSE values of 14.2 for the resin, 13.6 for the Colwell procedure, and 13.8, Bray 1 procedure (Table I).

The majority of relative yield data was greater than 80% of maximum (68% of observations), with very few points at the lower end of the scale. This would contribute to the poor relationship between relative yield and soil test P. High relative yields were due to the two high levels of fertilizer applied in 1992 at each site.

3.3. Field experiment 1995

Over the 6 sites used in 1995 and all fertilizer types, all tests showed a poor linear relationship between soil test P and level of applied P. The Bray 1 test gave the best relationship ($R^2 = 0.48$, n = 54), followed by Colwell procedure ($R^2 = 0.39$), and resin test ($R^2 = 0.27$) (data not presented). Multiple linear regression including site, fertilizer type, and level of P applied effects, showed a fertilizer type effect with a significantly different intercept ($P < 0.05$) for different fertilizer types, for all tests except resin. However, R^2 values for any test and fertilizer type were still poor with Bray 1 having the best relationship for RPR ($R^2 = 0.51$ n = 9).

Although the sites used for the 1995 evaluation were responsive to P ($P < 0.05$) and a range of soil P values and relative yield responses were represented, none of the tests adequately described the relationship between relative yield and soil test P (Fig. 5). All tests had similar RMSE values (23 for Bray 1, 24 for resin, and 25 for Colwell).

There was no improvement in the fit of the curves when each fertilizer was considered separately, with RMSE values ranging from 22 to 25 (Table II). Only when sites were considered separately, with or without considering each fertilizer type, was there any significant improvement. In general the Bray 1 test had the lowest RMSE values, particularly for the sites in southern Australia (V15, S24, and S25), which supported temperate legume and/or grass pastures. For the tropical sites, the resin test provided the best relationship for PAPR, while the Bray 1 test gave the best relationship for RPR.

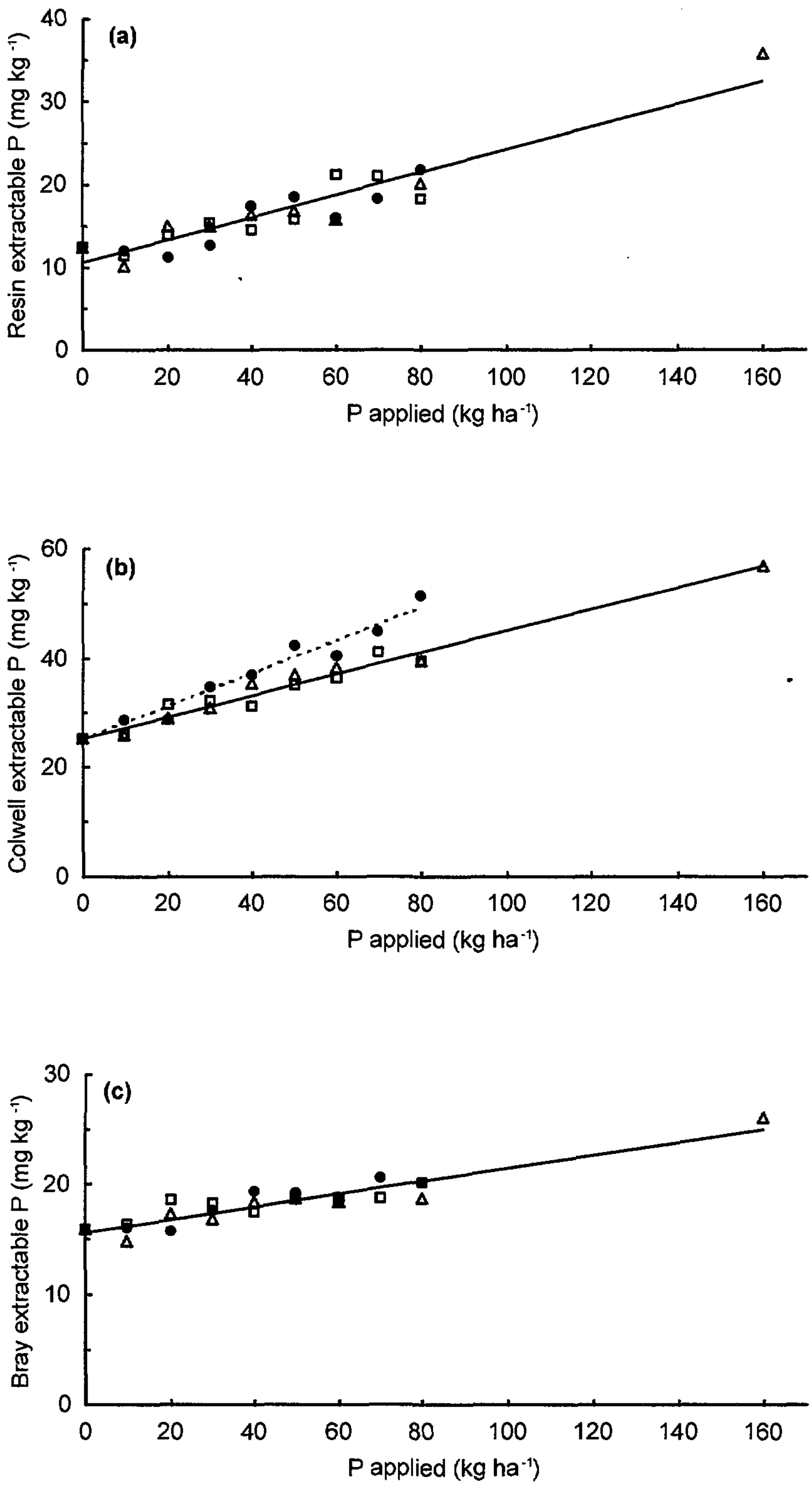

Fig. 1. The relationship between soil test P and the amount of fertilizer applied as TSP (●), PAPR (□), and North Carolina PR (Δ) in the glasshouse experiment for (a) the resin P: $Y = 10.53 + 0.137\,X$ ($R^2 = 0.87$); (b) the Colwell reagent: $Y = 25.5 + 0.296\,X$ ($R^2 = 0.96$) for TSP (-----), and $Y = 25.5 + 0.196\,X$ ($R^2 = 0.96$) for PAPR and North Carolina PR (————), and (c) the Bray 1 reagent: $Y = 15.7 + 0.058\,X$ ($R^2 = 0.86$) (reproduced with permission of CSIRO Publishing [10]).

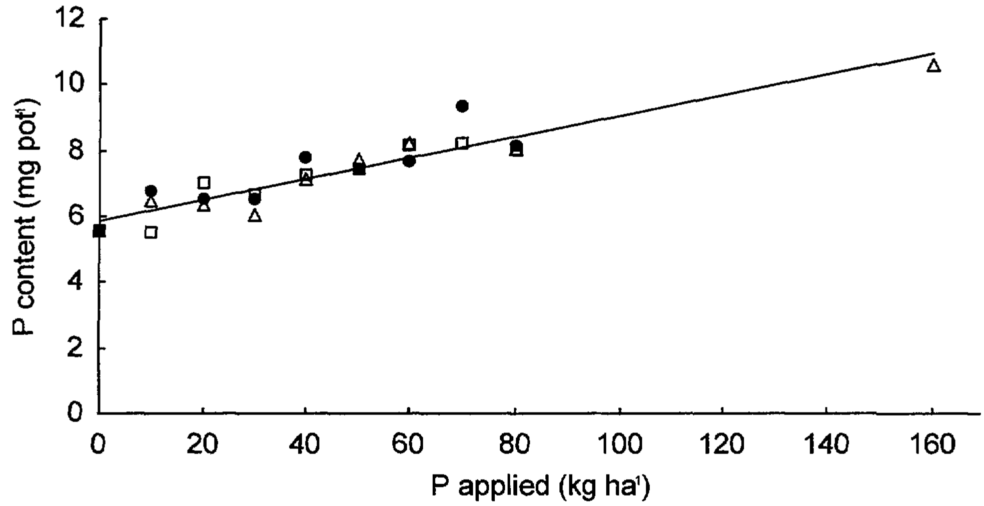

Fig. 2. The relationship between P content of Trikkala subterranean clover and the level of fertilizer applied as TSP (●), PAPR (□), and North Carolina PR (Δ) in the glasshouse experiment. The equation for the fitted line is: Y= 5.89 + 0.032 X (r^2 = 0.72) (reproduced with permission of CSIRO Publishing [10]).

TABLE I. SQUARE ROOT OF THE ERROR MEAN SQUARE (RMSE) VALUES[1] FOR FITTED MITSCHERLICH MODELS FOR THE RELATIONSHIP BETWEEN RELATIVE YIELD AND SOIL TEST P FOR RESIN, COLWELL REAGENT, AND BRAY REAGENT, OVER ALL FIELD SITES FOR TSP, PAPR, AND RPR IN 1993 (REPRODUCED WITH PERMISSION OF CSIRO PUBLISHING [10]).

Fertilizer		Soil test	
	Resin	Colwell	Bray 1
All fertilizers	19.0	16.8	16.5
TSP	21.9	21.9	17.8
PAPR	17.2	15.8	15.8
RPR	14.2	13.6	13.8

[1]The 'goodness' of fit for the relationship is indicated by a small RMSE. The range for the relative yield is 0-120%. The bigger the RMSE, the poorer the quality of the relationship between soil test P and relative yield.

4. DISCUSSION

All tests used in the field evaluation in 1993 and 1995 performed poorly when examined over a range of soil types and environments with very poor relationships between relative yields and soil test P (Figs. 4 and 5). In soil testing, it is assumed that the use of the relationship between relative yield (instead of absolute yield) and soil test P values minimizes seasonal variation. Our findings suggest that this is not so for the diverse soils and environments used in this study (Table I). The relationship between plant yield and soil test P are usually good where only one or a limited range of soils are used in glasshouse or field experiments [11-14]. Where evaluation of soil P tests has been performed at several sites under field conditions, relationships are often poorer than under glasshouse conditions, and the relationship between relative yield and soil test P differs depending on soil type, plant species or seasonal conditions [15-19].

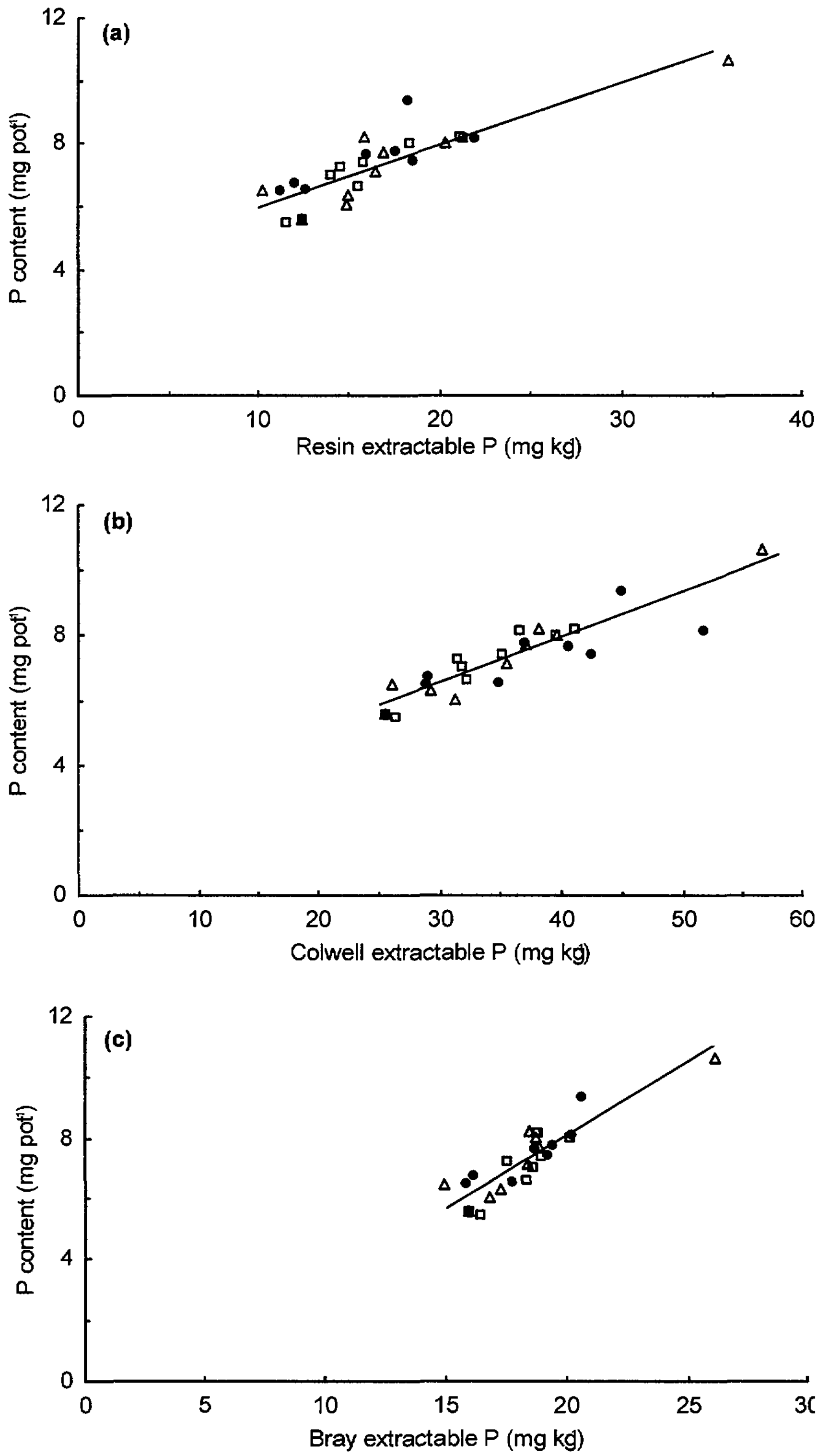

Fig. 3. The relationship for the glasshouse experiment between P content of Trikkala subterranean clover and the amount of (a) resin extractable P: Y = 3.96 + 2.0 X (R^2 = 0.72); (b) Colwell extractable P: Y = 2.38 + 0.14 X (R^2 = 0.82) and (c) Bray 1 extractable P: Y = -1.65 + 0.49 X (R^2 = 0.78). TSP (●), PAPR (□), North Carolina PR (Δ) (reproduced with permission of CSIRO Publishing [10]).

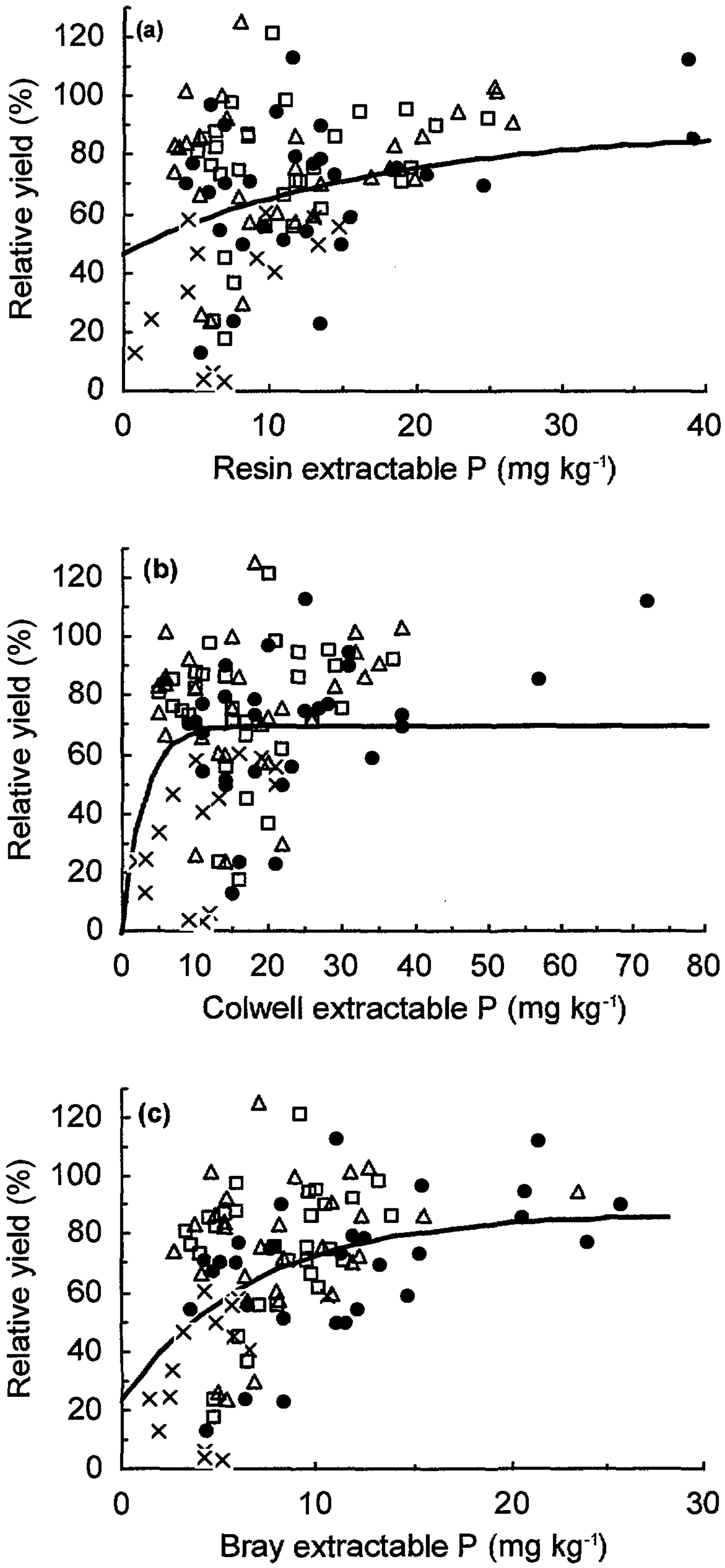

Fig. 4. The relationship between relative yield and soil test P for the 1993 field trial for (a) resin, (b) Colwell, and (c) Bray 1 with fitted Mitscherlich curves. TSP (●), PAPR (□), North Carolina PR (△) (reproduced with permission of CSIRO Publishing [10]).

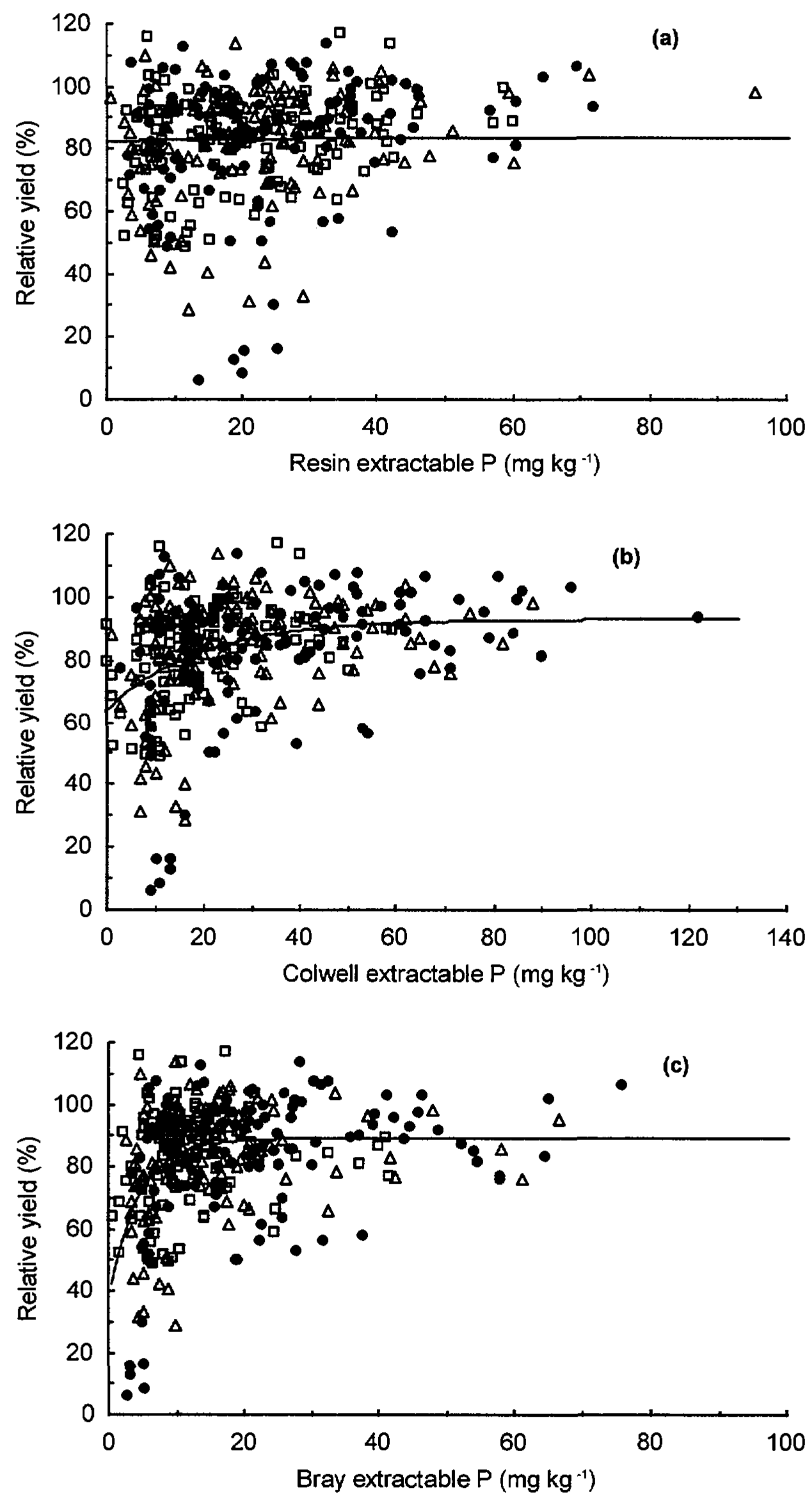

Fig. 5. The relationship between relative yield and soil test P for the 1995 field trial for (a) resin, (b) Colwell, and (c) Bray 1 soil P tests, with fitted Mitscherlich curves. TSP (●), PAPR (□), North Carolina PR (△) control (×) (reproduced with permission of CSIRO Publishing [10]).

TABLE II. SQUARE ROOT OF THE ERROR MEAN SQUARE (RMSE) VALUES[1] FOR FITTED MITSCHERLICH MODELS FOR THE RELATIONSHIP BETWEEN RELATIVE YIELD AND SOIL P TEST FOR RESIN, COLWELL REAGENT, AND BRAY REAGENT FOR TSP, PAPR, AND RPR FOR FIELD SITES IN 1995 (REPRODUCED WITH PERMISSION OF CSIRO PUBLISHING [10])

Site=	Q1 (Malanda)	Q2 (Tully)	Q3 (Mackay)	V15 (Hamilton)	S24 (Victor Harbour)	S25 (Kangaroo Island)	All sites
Fertilizer			RMSE				
				Resin			
All fertilizers	17.0	16.0	17.0	12.0	10.0	34.0	24.0
TSP	11.6	19.4	17.0	11.5	14.7	34.7	23.0
PAPR	16.2	15.3	13.7	14.9	9.6	36.8	22.0
RPR	16.2	19.9	18.9	10.9	9.5	40.4	24.0
				Colwell			
All fertilizers	23.0	12.0	19.0	13.0	12.0	34.0	25.0
TSP	18.4	9.4	13.7	12.5	15.8	34.8	22.0
PAPR	16.4	15.5	17.7	15.4	10.5	37.8	22.0
RPR	24.9	13.6	20.3	13.4	9.7	39.7	25.0
				Bray			
All fertilizers	16.0	11.0	18.0	11.0	10.0	32.0	23.0
TSP	12.2	10.6	16.7	10.9	11.5	31.7	23.0
PAPR	16.5	16.4	21.8	9.06	9.5	33.4	22.0
RPR	14.4	13.5	15.6	7.6	8.3	38.8	23.0

[1]The 'goodness' of fit for the relationship is indicated by a small RMSE. The range for the relative yield is 0-120%. As such the bigger the RMSE, the poorer the quality of the relationship between soil test P and relative yield.

There are a number of possible reasons why the relationship between relative yield and soil test P was not constant across the different sites in the present study. The first is that different seasonal conditions result in different relationships between relative yield and soil test values, even for the same species in the same soil, topdressed with the same P fertilizer [15, 20]. The second reason is the wide range of soil types that were used for this study, from a peat sand to a medium clay krasnozem soil [1]. Reuter *et al.* [19] were able to show that soil test P values related to 90% relative yield (critical values) for cereals changed with soil texture. The P sorption capacity of soils from sites in this national project was found to change with soil texture [21]. The third possible reason was the difference in botanical composition of the pastures across the sites. The sites contained tropical grass (site Q2), tropical legume (Q3), mixed perennial grass and clover (e.g. sites V15, and S24) and subterranean clover dominant sites at the remaining locations [1]. Bolland and Gilkes [20] and Reuter *et al.* [19] have shown how critical soil test values differ for different species. Different sites used for this study generally had different species present, and may subsequently have had different soil P levels for similar relative yields.

A fourth possible reason is due to the effect of the different types of P fertilizer. We were not able to show any differences in the Mitscherlich calibration curves for the field data in 1993 and 1995 between the different fertilizer forms because of the variable results in the data. However, differences in calibration relationships between WSP and RPR forms have been previously reported [22, 23]. The extent of RPR dissolution during a growing season, the P nutrition of the pasture, the ability of an empirical soil test to predict P availability, and the extent of RPR dissolution that occurs post-sampling all determines the efficacy of a soil test for RPR and the likelihood that there will be different calibration relationships for WSP and RPR fertilizer. The dissolution of RPR, and the availability of the dissolved P, depends on edaphic and climatic factors [21], which cannot be ascertained across sites by a single soil test. It is not surprising therefore that soil testing for RPR is being developed in conjunction with mechanistic models to describe RPR dissolution during the growing season [24, 25].

The results from the glasshouse and field experiments show that the resin test was not superior to either the Colwell or Bray 1 tests. On the contrary, the Bray 1 and to a lesser extent the Colwell test, were both slightly better than the resin test in the glasshouse study (Fig. 3). Similarly, there was a closer fit of relative yield data in the 1993 growing season to Bray 1 and Colwell extractable P than to the resin extractable P (Table I, Fig. 3), though all tests performed poorly. All soil tests were generally similar in their ability to predict relative yields from the P already present in the soil (Table II, Fig. 5).

The soil used for the glasshouse study was collected from site V12 in the National RPR Project. This soil was non-responsive to P in terms of clover yields, necessitating the use of P content data. The soil also had high levels of titrateable acidity [21]. These two factors meant there were limited dry matter responses to soil P test values, and conditions were very favorable for RPR dissolution, such that all soil tests were able to detect increases in available P in the RPR treatments (Fig. 1) and no fertilizer type was agronomically superior. These conditions contrast to those reported by Saggar *et al* [24] who found the resin test to be superior to the Olsen bicarbonate test where yield responses by perennial ryegrass to added P were recorded for all 4 soils used, and the WSP fertilizer was generally more effective than PR.

Only 2 levels of P were used in the field studies. A wider range of P levels would have provided a better defined soil P test calibration to evaluate the various soil tests procedures.

The resin test extracts many other elements simultaneously so it is a multi-element test. The test therefore has advantages in saving time and cost and when the analysis of other elements is considered [26]. The other soil P tests examined in this study were for P only.

5. CONCLUSION

In the glasshouse experiment, all soil P tests were reasonably effective in predicting P content responses to soil test P values for soils previously fertilized with 3 different fertilizer types. However, under field conditions all tests performed poorly when examined over a range of soil types and environments. On an individual site basis however, there were improved relationships between P applied and soil test P, and between relative yield and soil test P at some sites. This highlights the potential problems that all soil test procedures face when used over a wide range of soil types and environments.

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