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Methods of Measuring Taro Leaf Blight Severity and Its Effect on Yield

Fred E. Brooks, Plant Pathologist

ABSTRACT

Production of taro (*Colocasia esculenta*) in American Samoa depends on recently imported cultivars resistant to taro leaf blight disease (TLB). The purpose of this study was to find a practical, inexpensive way to monitor TLB and its possible effect on cultivar resistance and yield. Researchers commonly estimate resistance by percentage of plant surface damaged (disease severity) but this may not be the best measure for TLB nor relate to yield. Two trials were conducted at the American Samoa Community College Land Grant facility during 1999-2000 to assess resistance of taro cultivars P16 (Meltalt), P20 (Dirratengadik) and Rota (Antiguo). Disease severity was measured by two methods: a direct estimate of percent disease per leaf, and by the use of a pictorial key and rating scale. Other plant growth indicators measured included pseudostem diameter, plant height, number of leaves and suckers, corm weight and length of leaf life. The direct estimate of disease and pictorial key were strongly correlated in both trials but direct estimates were significantly higher than estimates made with the pictorial key. This may have been due to intra-rater repeatability or inter-rater reliability. There was no correlation between disease severity and yield (corm weight) with either method. Plant height and number of leaves and suckers per plant were the best indicators of yield. These indicators are easy to assess in the field but may vary between cultivars.

INTRODUCTION

The purpose of this study was to find a practical, inexpensive method of monitoring taro leaf blight disease (TLB) and its effect on cultivar resistance and yield. In American Samoa, production of taro, *Colocasia esculenta* (L.) Schott, was devastated by an epidemic of TLB in late 1993-1994 (Trujillo et al. 1997). Taro production fell from 357,000 kg (786,000 lb) per year before the epidemic to less than 5,000 kg (11,000 lb) by the end of 1994 (Economic Development and Planning Office 1994).

During 1994-1995, taro cultivars from the American Pacific were tested in Hawai'i (Greenough et al. 1996) and Guam (Wall & Weicko 1998) for resistance to the TLB fungus, *Phytophthora colocasiae* Racib. The most promising cultivars from the Hawai'i screening were sent to American Samoa in 1997. Farmers are now growing resistant cultivars in fields that have been without taro (*C. esculenta*) since the epidemic.

Researchers use several methods to measure disease severity. Most are based on the amount of a plant part (leaf, stem, fruit, etc.) affected by disease. A graph of disease severity over time (disease progress curve) is a direct measure of disease progress, an indirect measure of the pathogen population, and is usually related to yield (Fry 1982). The disease progress curve can affect our choice of management strategies, monitor their effectiveness, and permit disease forecasting (Fry 1982).

Taro leaf blight severity is usually measured by estimating the percent disease per leaf, finding an average for the plant, then calculating a mean disease rating for the cultivar (Gollifer & Brown 1974, Jackson et al. 1980, Semisi et al. 1998). Cox (1986) and Cox and Kasimani (1990a) discussed the limitations of these methods, pointing out that since TLB is most severe on lower leaves, the more leaves a plant retains, the higher its disease rating (percent disease). Plants that lose most of their leaves to TLB (i.e. severely diseased) will usually have a lower disease rating. Cox and Kasimani (1990) therefore recommend counting leaves, or total healthy leaf area, to assess TLB severity and not measuring percent disease.

Measuring TLB severity by percent disease may not always relate to yield (Cox 1986). Gollifer and Brown (1974) found no correlation between percent disease and yield but did find a positive correlation between disease severity and number of leaves per plant and leaves per plant and corm yield.

American Samoa does not have a taro breeding program at present and depends on the resistance of newly introduced cultivars for taro production. One goal of the American Samoa Community College Land Grant Program (ASCC) is to monitor this resistance. The objective of this study was to evaluate two commonly used methods of assessing disease severity, several plant growth parameters, and their relationship to TLB resistance and yield.

MATERIALS AND METHODS

SITE PREPARATION AND PLANTING

Two six-month trials were conducted at the ASCC Land Grant facility on the main island of Tutuila, American Samoa. The first trial began during the dry season, 21 June, and ended in the wet season, 27 December 1999. The second trial was planted in the wet season, 14 February, and harvested during the dry season, 22 August 2000. Plant spacing and trial design for these studies were affected by a shortage of available land. Plots were randomized with no repetitions. Main plots consisted of resistant cultivars interplanted among a suspected susceptible cultivar. Subplots consisted of resistant cultivars only; a susceptible cultivar only plot was added to the second trial. Main plots measured 6.75 x 3 m, subplots 3.75 x 3 m with 1 m between plots. In main plots, four three-plant rows of resistant taro alternated with four three-plant rows of susceptible taro, producing 12 data plants of each cultivar: 26 TLB-resistant border plants surrounded each main plot. Subplots each contained 12 data plants surrounded by 18 border plants. Three additional subplots, one for each cultivar, were established for comparison with test plots. These control plots were treated every 14 days with a 15-20 cm banded soil application of metalaxyl (Ridomil 2E, Ciba-Geigy, Greensboro, NC), at 8 ml a.i. per plot. Metalaxyl is a fungicide with systemic activity against *Phytophthora* species.

TARO CULTIVARS

Taro leaf blight resistant cultivars P16 (Meltalt) and P20 (Dirratengadik) from the Republic of Palau were selected as a result of Hawai'i field trials (Greenough et al. 1996, Trujillo et al. 1997) and discussions with local taro farmers. Antiguo, renamed Rota in American Samoa (P. Gurr, pers. comm.), was used as the susceptible cultivar in these trials. In the Hawai'i taro trials average leaf damage per cultivar was 8% for P16 and P20 and 28% for Antiguo (Rota) (Greenough et al. 1996, Trujillo et al. 1997). All plants were grown in the ASCC multiplication plot and naturally infected with TLB. Planting material from this plot consisted of leaf petiole bases (pseudostems) with part of the corm attached (Samoan = *tiapula*) and provided a natural source of trial inoculum (Cox & Kasimani 1988, Hicks 1967, Semisi et al. 1998). Optimum length of harvest for different Palauan cultivars in American Samoa has not been determined but most farmers suggest between six and nine months. We harvested data plants at six months, measured leaf base (tiapula) diameters and weighed corms after removing roots and soil.

DISEASE SEVERITY.

We measured taro leaf blight severity from the onset of disease in the field and continued at two-week intervals until harvest. Disease severity was defined as percent of plant leaf surface affected by TLB, either lesions or lesions plus lesion-related chlorosis and yellowing (James 1971). New, partially furled leaves and old leaves touching the ground were not evaluated. Two assessment methods were compared: a direct estimate of percent disease and a pictorial key with a pretransformed rating scale of 0-6.

Direct estimate (Method 1). Disease severity was estimated directly for each leaf of a data plant; 0 = no disease, followed by increments of 5, 10, 25, 50, 75, 90 and 100% disease. If disease severity was between two increments, we recorded the higher increment during the first trial; estimates were rounded to the nearest increment during the second trial. Percent estimates of disease severity were converted by angular transformation before statistical analysis (Little & Hills 1978). To calculate disease severity and number of healthy leaves per plant:

DS = å %TLB / Lvs

HLvs = (100% - DS / 100) x Lvs

where,

DS = disease severity (percentage)

%TLB = estimated percentage taro leaf blight per leaf

Lvs = number of leaves per plant

HLvs = effective number of healthy leaves per plant

Pictorial key (Method 2). A 7-point rating scale was created by angular transformation, according to the method of Little and Hills (1978). Disease severity was estimated by comparing TLB damage on each leaf with a modified pictorial key (Gollifer & Brown 1974). We recorded the rating under the picture most closely matching leaf damage, from 0 = no disease to 6 = more than 93% diseased (Figure 1*)*. All data analysis for disease severity was done on the transformed ratings then back transformed to obtain percent values (Little & Hills 1978). To calculate disease severity and number of healthy leaves per plant:

$DRP = \sum DRL / Lvs$

$HLvs = 6 - DRP \rightarrow BT\% \times Lvs$

Note: $DS = DRP \rightarrow BT\%$

where,
DRP = estimated disease rating per plant (0-6)
DRL = estimated disease rating per leaf
Lvs = number of leaves per plant
DS = disease severity (percentage)
BT% = rating back transformed to a percentage
HLvs = effective number of healthy leaves per plant

LEAF LIFE MEASUREMENT.

Leaf life of each cultivar was measured during the first trial, beginning on day 140, by tagging one emerging leaf on each data plant. An emerging leaf was defined as one with petiole showing and some or all of the leaf still furled. Labeled leaves were assessed every-other-day for six weeks and the date recorded when the leaf surface was covered with TLB lesions, chlorosis and yellowing associated with lesions, or both. Average leaf life, in days, for each treatment and cultivar was calculated. The leaf life study was not repeated during the second trial.

TIAPULA DIAMETER, PLANT HEIGHT, AND NUMBER OF LEAVES AND SUCKERS.

To evaluate the relationship between the size of *tiapula* planted, average increase in diameter, and yield (corm weight), *tiapula* diameters were measured directly below petiole bases at planting and harvest. Plant height was measured and the suckers and leaves counted on each of eight assessment days, beginning with the onset of disease. The effective number of healthy leaves per plant was also calculated using the disease severity formulas. New leaves were not counted unless they were unfurled nor were old leaves with collapsed petioles (leaf blade touching the ground).

DATA ANALYSIS.

Differences in taro leaf blight disease severity, plant height, increase in *tiapula* diameter, number of leaves and suckers, average leaf life, and corm weight for main plot and subplot treatments for each cultivar were compared by one-way analysis of variance. Means separations, when appropriate ($P < 0.05$), were made with Tukey's test for pairwise comparisons. Linear correlations between all methods and plant growth parameters were compared by Pearson's product-moment correlation ($P < 0.05$) (SigmaStat, SPSS Inc., Chicago, IL).

RESULTS

DISEASE SEVERITY

There was a strong positive correlation in both trials between Method 1, the direct estimate of disease severity, and method 2, use of a pretransformed pictorial key (Figure 2). The correlation was higher in the first trial ($r^2 = 0.95$) than the second trial ($r^2 = 0.52$). Method 1 mean estimates were significantly higher ($P<0.001$) than Method 2 estimates in both trials (Tables 1, 2).

Severity of TLB in the first trial (average of untreated plots) was 11%, 14%, and 13% for P16, P20 and Rota, respectively (Method 1). For the second trial, disease severity was 10% for P16, 8% for P20, and 9% for Rota. Disease in fungicide-treated plots was lower than in untreated plots, but only significant in the first trial (Tables 1, 2).

PLANT HEIGHT

Taro cultivars P20 and Rota were significantly taller ($P < 0.05$) in the first trial than in the second trial (Tables 1, 2). During the first trial, all three cultivars were significantly shorter in the treated plots at the poorly drained lower end of the field than in the untreated plots at the upper end of the field (Table 1). There was a strong positive correlation in both trials between plant height and corm weight ($r^2 > 0.50$) in 16 of 19 plots (Figure 3).

TIAPULA DIAMETER

There was no correlation between the diameter of *tiapula* planted and final corm weight. However, there was a positive linear correlation between the increase in *tiapula* diameter and corm weight ($r^2 = 0.25$) in five of nine plots in the first trial. In the second trial, three of 10 plots had a positive correlation greater than $r^2 = 0.25$; three plots, however, had a negative correlation between *tiapula* diameter increase and corm weight. Average increase in *tiapula* diameters for P20 and Rota was significantly greater in the first trial than in the second ($P < 0.001$).

NUMBER OF LEAVES

There was a strong positive correlation ($r^2 = 0.85$) between the number of leaves per plant counted during assessments and the effective number of healthy leaves calculated by formula. The number of leaves on plants in plots treated with metalaxyl was greater in both trials than the number of leaves in untreated plots for all cultivars (Tables 1, 2). Eight of the 13 plots not treated with fungicide in the two trials demonstrated a moderate positive correlation ($r^2 > 0.25$ but < 0.50) between the number of leaves and corm weight; two plots showed a high correlation ($r^2 > 0.50$). Average leaf number per plant was highest in both trials at the beginning of assessments, decreased, then stabilized (Figure 3).

NUMBER OF SUCKERS

Differences between the average number of suckers per plot were not significant for most treatments. In the first trial, however, plants in the slow draining P16-treated plot produced significantly fewer suckers than P16 plots in either trial ($P < 0.001$). Plants in this plot were also significantly shorter and had the smallest corm weights (Table 1). Seven of 19 plots indicated a moderate positive correlation between the number of suckers per plant and corm weight ($r^2 > 0.25$ but < 0.50) and in three plots the correlation was high ($r^2 > 0.50$).

AVERAGE LEAF LIFE

Tagged leaves of P20 lived longest (35 days), followed by P16 (32 days) and Rota (29 days). In the first trial, tagged leaves lived significantly longer in fungicide-treated plots of cultivars P20 and Rota than in untreated plots. There was no correlation between leaf life and corm weight for either P16 or Rota. There was, however, a positive correlation between leaf life and corm weight in plots of P20-only ($r^2 = 0.36$) and P20 treated with metalaxyl ($r^2 = 0.41$). Leaf life was not measured directly during the second trial.

CORM WEIGHT

Average corm weights for P20 and Rota, all treatments, were significantly greater in the first trial than the second ($P < 0.001$). There was no correlation between disease severity and corm weight for any cultivar in the first trial and only a moderate positive correlation in two plots during the second trial ($r^2 = 0.30$). Though there were no consistent treatment effects on corm weight (Tables 1, 2), there was a good positive correlation between leaf number, number of suckers, plant height, and corm weight.

DISCUSSION

There was a strong positive correlation in the first trial between disease assessment Method 1, the direct estimate of disease per leaf, and Method 2, the pictorial key. However, the average difference in estimates of TLB severity for P16, P20 and Rota were significantly higher using Method 1¾6%, 7% and 7% higher, respectively (Figure 2). This was partly due to a tendency to "round up" with Method 1 and to round up or down with Method 2. For example, if leaf area affected by TLB was estimated to be slightly above 25%, it was given a rating of 50% (25%-50%) using Method 1. With Method 2, however, the picture/rating most closely matching the affected leaf may have been selected and rated "2" (8-25%); when back transformed, "2" would be 25%. In the second trial, we rounded estimates for both methods to the closest increment. Though the average difference between the methods was smaller¾4%, 2% and 2% higher for P16, P20 and Rota, respectively¾TLB severity still measured significantly higher with Method 1 than Method 2 in over half the plots (n = 19).

Intra-rater repeatability and inter-rater reliability were not tested before these trials and offer another possible source of assessment error. The former is a measure of the consistency of repeated assessments by a worker in a given area, the latter a measure of agreement between the disease assessments of different workers for the same area (Nutter 1993). Finally, when taro leaf blight severity was assessed with the pictorial key, workers were inclined to estimate leaf blight damage by percent and transpose it to a rating number rather than match the pictures in the key to leaf damage. Workers not able to use, or not taught, a direct estimate of disease severity, may use the pictorial key more effectively.

In our trials, there was no correlation between disease severity measured as percent disease and yield (corm weight). This disagrees with typical plant disease evaluations in which disease severity and yield are directly related (Fry 1982). It is in agreement, however, with Cox (1986), who stated that methods based on percent disease ratings of available leaves only, were "trivial". Healthier, more resistant plants tend to maintain heavily diseased leaves while susceptible plants lose diseased leaves, retaining only young leaves with fewer infections. Our results also support those of Gollifer and Brown (1974), who found no correlation between percent disease and yield but did find a positive correlation between leaves per plant and corm weight. The lack of correlation between percent disease and yield in our trials may have been from either a small reduction in yield at these low disease levels, too small a difference between treated and untreated plots, or both. Finally, though rare and difficult to quantify, these cultivars may be exhibiting resistance in a broad sense, one that includes tolerance, or the ability to maintain high yields as disease severity increases (Fry 1982, Schafer 1971).

Plant height and number of suckers per plant were better indicators of corm weight than disease severity in these trials. Plant height had a medium to high correlation with corm weight in 17 of 19 plots; the number of suckers and corm weight were positively correlated in nine of 19 plots. Plant height, number of suckers and corm weight were highest in untreated plots at the upper end of the field in the first trial and lowest in treated plots at the poorly drained lower end (Table 1). Fungicide-protected plants in both trials had less taro leaf blight damage, more leaves, and longer leaf life, yet were the shortest and tended to have lower corm weights. These results, along with field observations, suggest conditions in the field may have more effect on yield at low disease levels than TLB.

Some local farmers believe suckers take energy from the mother plant, reducing corm size. Results of this study indicated the number of suckers produced by a plant had no significant negative effect on corm weight. Initially, suckers were a nutrient sink with the mother plant acting as the nutrient source. When suckers form leaves with complete photosynthetic ability, they synthesize their own nutrients (Salisbury and Ross 1992). The initial loss of nutrients from the mother plant to produce suckers may be offset by an overall increase in photosynthesis stimulated by these nutrient sinks. A study by Gifford and Evans (1981) showed the removal of potato tubers causes a marked reduction in plant photosynthesis. The number of suckers produced by the mother taro plant, however, may just be another indicator of plant health and healthy, vigorous plants tended to be larger and produce larger corms.

There was a good correlation (r^2>.25) between the number of leaves per plant and corm weight in plots not treated with fungicide but the relationship between leaf life and corm weight was inconsistent. One reason for these inconsistencies may have been conditions in the field, amended in 1998 with 15 truckloads of volcanic cinders and undeveloped subsoil high in clay. During the first trial, the lower end of the field was routinely flooded after moderate to heavy rainfall. Treated plants located at the lower end of the field (windward) to reduce interplot interference (Paysour & Fry 1983) were often waterlogged and never as vigorous as plants in the better-drained, upper part of the field. They also had below-average corm weights. Treated plots were randomly distributed in the second trial and corm weights were average for these plots. The strong correlation between number of assessed leaves per plant and number of healthy leaves (calculated) was to be expected at low disease levels. Simple leaf counts in the field were a fast, effective measure of disease severity and yield but may not remain so at higher disease levels.

We measured *tiapula* diameters at the base of leaf petioles at planting with the assumption that large *tiapula* would produce larger corms than small *tiapula*. There was no correlation, however, between initial tiapula diameters and final corm weight. One possible explanation is that conditions in the field during the trial moderated the initial variability in *tiapula* size. Another possibility is the difference in diameters, 13-37 mm, was not great enough to influence final corm weight. The correlation between average increase in tiapula diameters and corm weight was also inconsistent: eight of 19 treatments showed a positive correlation but three were negatively correlated.

Plant height, number of suckers, and number of leaves per plant were better indicators of yield than severity of taro leaf blight disease. These measurements were fast, easy to perform and the most consistent indicators of yield. They may vary, however, between cultivars. P20, for example, produced the tallest plants and largest corms but developed few suckers. P16 was short with smaller corms but produced more leaves and suckers. Rota was almost as tall as P20, produced slightly fewer suckers than P16 and developed small corms. At disease levels near 10%, conditions in the field appeared to have a greater effect on corm size than TLB and recommendations to growers will continue to emphasize maintaining plant vigor. Future research on taro leaf blight assessments will include known susceptible cultivars and tests of intra-rater repeatability and inter-rater reliability.

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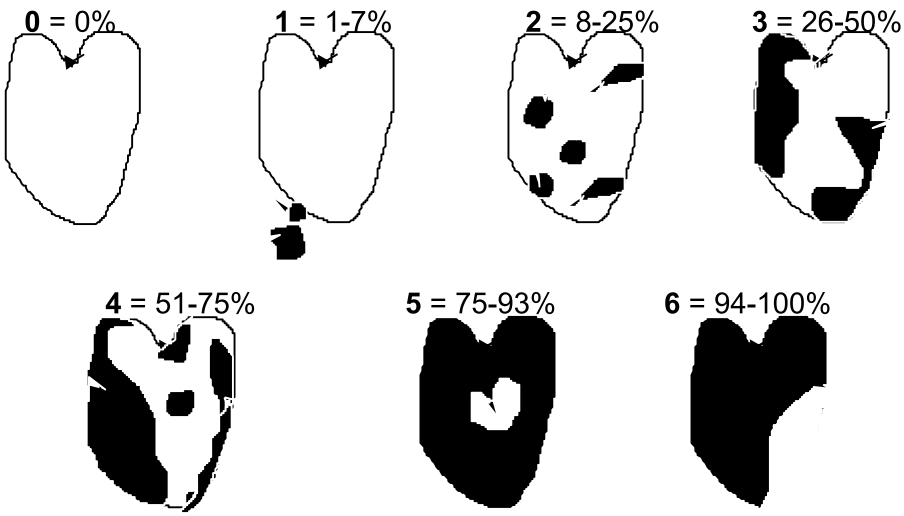

Figure 1. Pretransformed pictorial key for assessing taro leaf blight disease (Method 2) in American Samoa taro trials, 1999-2000. Pictorial key modified from Gollifer and Brown (1974); pretransformed rating scale from Little and Hills (1978).

Table 1. Pairwise comparison of three taro cultivars in a trial conducted at the ASCC Land Grant facility, 21 June to 27 December 1999. Each entry represents 12 data plants either in separate plots (-only), interplanted (+Rota), or treated with metalaxyl (-treated).

CULTIVAR	**Height**[1](cm)	**Leaves**[2](no.)	**Suckers**(no.)	**Stem Dia**(cm)	**Corm Wt.**(g)	**Method 1**	**Method 2**
P16-only	71.2a	4.9a	6.9a	2.7a	238.7a	12.5a	5.5a
P16+Rota	66.4a	4.8a	6.3a	2.5ab	216.1ab	9.0a	4.5a
P16-treated	48.4b	5.6b	2.0b	2.1b	155.2b	5.5b	1.0b
P20-only	99.4ab	4.4a	4.3a	3.8ab	484.1a	13.5a	7.0a
P20+Rota	106a	4.3a	5.8a	3.5a	576.3a	14.5a	8.0a
P20-treated	89.9b	4.9b	4.3a	4.5b	417.4a	6.5b	2.5b
Rota+P20	100.3a	4.1a	7.1a	2.7a	351.5a	13.5a	7.0a
Rota+P16	84.6b	4.7b	5.6b	4.2b	224.3a	11.8a	5.5a
Rota-treated	78.1b	5.1b	5.9b	3.0a	281.5a	6.5b	1.5b

Table 2. Pairwise comparison of three taro cultivars in a trial conducted at the ASCC Land Grant facility, 14 February to 22 August 2000. Each entry represents 12 data plants either in separate plots (-only), interplanted (+Rota), or treated with metalaxyl (-treated).

CULTIVAR	**Height**[1] (cm)		**Leaves**[2] (no.)	**Suckers** (no.)	**Stem Dia.** (cm)		**Corm Wt.**
(g)	**Method 1** (% disease)		**Method 2** (% disease)				
P16-only	65.0a	4.9a	7.3a	2.2a	246.2a	10.0a	5.6a
P16+Rota	63.4a	4.8a	8.3a	2.0a	189.2b	9.3a	6.2a
P16-treated	60.8a	5.8b	7.8a	2.6a	229.7ab	6.5b	3.9a
P20-only	65.4a	3.9a	1.0a	2.7a	216.0a	6.7a	5.5ab
P20+Rota	79.0b	4.0a	4.0b	2.8a	339.0b	8.9a	7.3b
P20-treated	65.7a	4.5b	2.5ab	2.7a	280.3ab	5.2a	2.5a
Rota-only	60.0a	4.0a	5.5a	1.7a	113.3a	7.6a	8.1a
Rota+P16	64.5a	3.6a	5.5a	1.9a	109.8a	9.8b	6.5a
Rota+P20	68.8a	3.9a	6.0a	2.1a	146.0a	9.2b	6.2a
Rota-treated	62.0a	4.0a	5.0a	1.9a	129.6a	6.2a	3.4a

[1] Differences between means within each column followed by the same letter are not significantly different (Tukey test, $P<0.05$). Comparisons are within each cultivar only and not between cultivars.

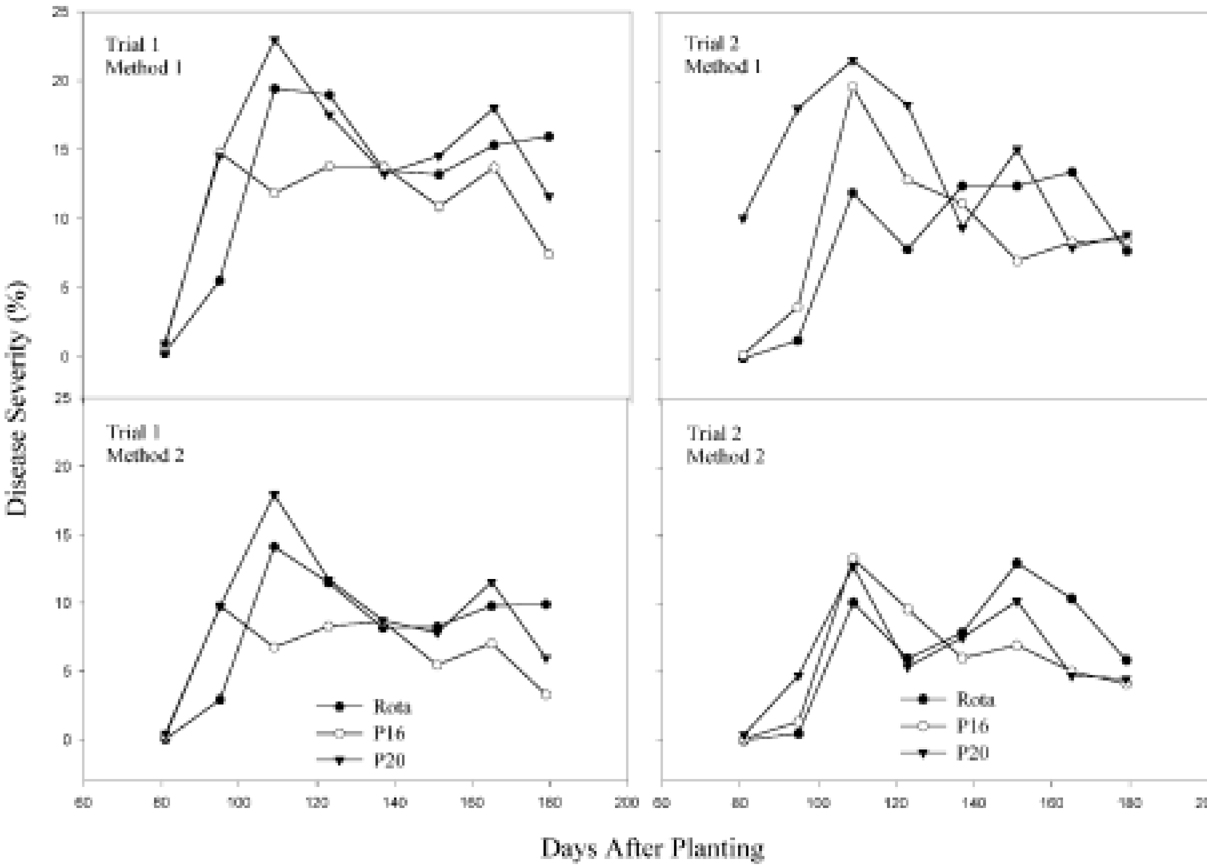

Figure 2. Comparison of two methods assessing taro leaf blight severity during trials at ASCC Land Grant facility, Tutuila, American Samoa. Trial 1 (left side) was planted on 21 June and harvested 27 December 1999. Trial 2 (right side) was planted 14 February and harvested 22 August 2000. Upper graphs represent a direct estimate of percent disease (Method 1); lower graphs illustrate use of a pictorial key (Method 2). Each data point represents plants in plots not treated with fungicide (n = 24 or 36).

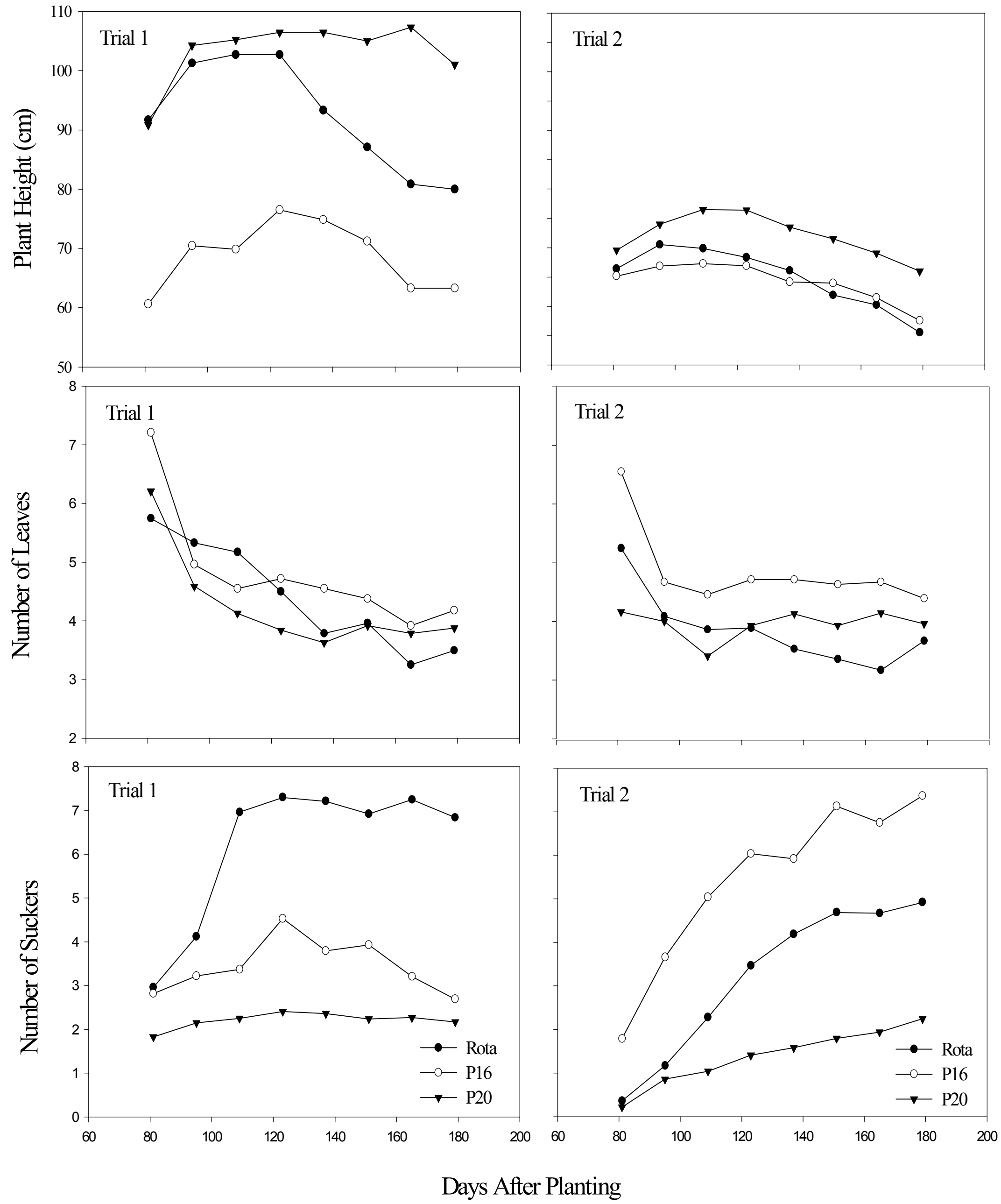

Figure 3. Comparison of average plant height, number of leaves (assessed), and number of suckers during six-month taro trials at ASCC Land Grant facility, American Samoa: Trial 1 (left) 21 June to 27 December 1999; Trial 2 (right), 14 February to 22 August 2000. Each data point represents at least 24 plants. Plots were assessed every two weeks, beginning 80 days after planting and continuing until harvest.