METHODS FOR DISEASE ASSESSMENT IN APPLES, INCLUDING GROWTH STAGES, LEAF EMERGENCE, BLACK SPOT AND POWDERY MILDEW

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ABSTRACT

Non-destructive sampling protocols are described for simultaneous assessment of disease incidence, growth stage and leaf emergence in apple trees for use in disease management, epidemiological and host-disease interaction studies on black spot (*Venturia inaequalis*) and powdery mildew (*Podosphaera leucotricha*). A new decimal code for apple growth stages from dormancy to fruit set is described. Assessments are carried out on 10 randomly selected shoots of specified type per tree on each of 4-6 trees per experimental plot or orchard block. 50-100 fruit per tree are assessed for incidence of black spot and russet. A minimum of two assessments are recommended, one in early December, and one pre-harvest.

Keywords: apple disease assessment, *Venturia inaequalis, Podosphaera leucotricha*, decimal growth stages, leaf emergence

INTRODUCTION

Apple disease management has usually focused on control requirements for diseases individually, and disease assessment methods have reflected this, eg. Croxall *et al.* (1952a,b). The need for greater integration of various components of orchard management to improve production efficiency and sustainability requires a research approach which explicitly addresses interactions between the host plant and disease. Important aspects of disease management which depend on the host plant include the scheduling of pesticides according to growth stages (even when weather-based forecasting systems are used, eg. Beresford and Spink 1992; van der Zwet *et al.* 1994), and the effects of leaf emergence on the susceptibility of trees to disease. The latter arises because apple leaves are most susceptible to black spot and powdery mildew infection while they are expanding (Szkolnik 1978; Butt *et al.* 1990) and consequently the seasonal timing of leaf emergence is of obvious importance in optimising fungicide use.

This paper describes non-destructive assessment methods developed in New Zealand from 1988-1995 for sampling of black spot and powdery mildew, and simultaneous collection of host development (growth stage) and leaf production data.

RECORDING METHODS

A recording sheet was developed for disease and host assessment (Table 1). It allows collection of data on incidence of black spot and powdery mildew on leaves, black spot and russet on fruit, numbers of leaves per shoot, numbers of fruit per cluster, and the incidence of shoots with powdery mildew infection of the growing apex.

Table 2 contains a decimal code describing principal and secondary growth stages for apples and gives an example of how the growth stages of three cultivars would be interpreted on one date during the flowering period. The rationale for the development and use of these assessment methods is described below.

TABLE 1: Example of a recording sheet for leaf and fruit disease, number of leaves per shoot and number of fruit per cluster. Italic type indicates assessment results for the leaves on 10 terminal extension shoots and the fruit on 10 fruiting spurs on one tree.

SAMPLE DATE: *7 December 1994* **CULTIVAR** *Fuji*
PLOT NUMBER: *12* **TREE NUMBER:** *3*

	LEAVES SHOOT TYPE *Terminal extension*				FRUIT SHOOT TYPE *Spur on 2nd-year wood*			
Shoot/ cluster #	No. lvs	B'spot on lvs	Mildew on lvs	Apical mildew	No. fruit	B'spot on fruit	Russet cat. 1	Russet cat. 2
1	*7*	*0*	*0*	*0*	*3*	*0*	*0*	*1*
2	*8*	*1*	*0*	*0*	*5*	*2*	*0*	*0*
3	*[1]	*0*	*0*	*0*	*4*	*0*	*1*	*0*
4	*	*0*	*0*	*0*	*2*	*0*	*0*	*0*
5	*	*0*	*2*	*0*	*5*	*0*	*1*	*2*
6	*	-[2]	-	*1*	*3*	*1*	*0*	*0*
7	*	*2*	*0*	*0*	*5*	*0*	*2*	*0*
8	*	*0*	*0*	*0*	*2*	*1*	*0*	*0*
9	*	*0*	*1*	*0*	*2*	*0*	*2*	*0*
10	*	*1*	*0*	*0*	*4*	*2*	*1*	*0*

[1] Number of leaves not counted on shoots 3-10 (see text)
[2] Apical powdery mildew precludes measurement of other variables

TABLE 2: Decimal growth stages for apples from dormant to fruitlet formation, and an example of proportion of shoots or flower clusters in each stage for four cultivars assessed on 19 October 1994. Mean growth stages were calculated to be: Braeburn, 3.0; Fuji, 2.2; Granny Smith, 2.3; Royal Gala, 2.1.

Principal growth stage		Secondary growth stage		Proportion of shoots or clusters Braeburn	Fuji	Granny Smith	Royal Gala
0	Breaking of dormancy	**0.0**	dormant				
		0.3	buds swollen				
		0.7	bud scales opening				
1	Bud burst	**1.0**	First leaves visible				
		1.3	Green tip				
		1.7	Centimetre green				*0.10*
2	Flower emergence	**2.0**	Tight cluster		*0.60*	*0.40*	*0.50*
		2.3	Open cluster		*0.20*	*0.30*	*0.30*
		2.7	Pink (less than 50% of flowers open per cluster)	*0.10*	*0.20*	*0.20*	*0.10*
3	Bloom	**3.0**	Full bloom (at least 50% of flowers with anthers visible per cluster	*0.80*		*0.10*	
		3.3	30% petal fall	*0.10*			
		3.7	70% petal fall				
4	Fruitlet formation	**4.0**	Fruitlet beginning to swell				

DISCUSSION

Growth stage assessments

Apple development has distinctive stages from bud burst to fruit set and these are the basis for conventional growth stage assessments. Terms used by the New Zealand Fruitgrowers Federation (Anon 1978) are followed in this paper: dormant, bud movement, bud burst, green tip, centimetre green, cluster, pink, full bloom, petal fall and fruitlet. We propose that these stages be incorporated into a decimal growth stage code for apples, following the principles successfully used for cereal growth stages (Zadoks *et al.* 1974). This allows numerical treatment of growth stages, including averaging and interpolation between stages. Five principal growth stages are proposed from dormancy to fruit set:

0) breaking of dormancy
1) bud burst
2) flower emergence
3) bloom
4) fruitlet formation

Secondary stages within each of these principal stages are described on a 0-9 scale (Table 2).

The decimal growth stage codes, which apply to individual buds, shoots, or flower or fruit clusters, can be written in a shorthand form, eg. GS 3.5 (half petal fall). The mean growth stage of a tree or orchard block can be determined by assessing the proportion of shoots at each stage and summing of the products of each code value and proportion. When averaging categorical data like this, attention must be given to whether or not the data are normally distributed. The distribution can be visualized when growth stages are recorded as in Table 2, and a range should be included when quoting a mean growth stage, eg. GS 3.5 (2.7-3.7).

A decimal code for both annual and perennial crops was proposed by Lancashire *et al.* (1991) which included both vegetative and reproductive parts of the plant in one system. Their sequence of principal growth stages dealt first with vegetative development (stages 0-4), then reproductive development (stages 5-8). For apples trees, reproductive development is concurrent with vegetative development and the numerical sequence of Lancashire's growth stages was therefore not biologically meaningful for apples. We use the decimal code for reproductive development only, and assess vegetative development (including leaves produced on fruiting shoots) as numbers of leaves which have emerged, leaf area or shoot length (see below). This produces true continuous variables which are preferable to descriptive stages. Before flower parts become visible at GS 2.0, flower buds cannot be distinguished from vegetative buds and therefore early "reproductive" growth stages actually describe the appearance of buds and leaves which are non-reproductive parts.

From fruit set to ripening there are few visually distinctive stages and reproductive development is characterized by increasing fruit size and colour changes. Growth stages after fruit set require further study and are not included in the current paper.

Shoot types and numbers of leaves

Leaves are produced on various types of apple shoots and the following type descriptions broadly follow Abbot (1984):

1) Terminal extension shoots, which grow for most of the season until the terminal bud has formed.
2) Lateral extension shoots, which grow for most of the season, but are less vigorous than terminal shoots.
3) Non-fruiting spurs, which break bud later than fruiting spurs and produce leaves for a limited period, giving rise to a rosette of leaves with a terminal bud at their centre.
4) Bourse shoots, which arise from vegetative buds in leaf axils on the flower-bearing shoot or bourse.
5) Fruiting spurs, which produce a rosette of about seven leaves, then a cluster of flowers which develop into fruit. The leaves remain for most of the season.

When assessing numbers of leaves in conjunction with disease, leaves are counted downwards from the top, and number one is the youngest fully unrolled leaf. Small leaves at the base of the shoot, which were the first to emerge from the bud, tend to be shed early in the season and are not usually assessed. The variability in the number of leaves present on a given shoot type at a given date is small compared to the variability in the incidence of disease on those leaves. It is therefore unnecessary to count all the leaves on every shoot on which disease is assessed. In Table 1, for example, the number of leaves has been counted for two out of the 10 shoots assessed for disease.

Disease assessment

Disease can be measured as incidence (percentage of leaves or fruit in a sample with disease), or as intensity or severity (percentage area of leaves or fruit diseased, or number of lesions or colonies per leaf or fruit). For black spot on leaves, incidence and severity are correlated, and the same is true for incidence and severity on fruit (Jeger 1981). As the time required for assessment is important, and as incidence can generally be determined more quickly than severity, our methods use disease incidence. Although Croxall *et al.* (1952a) developed a rapid method for black spot severity measurements, their method was quite subjective, involving visual assessment of whole branches in relation to a key. Incidence is an appropriate measure for black spot on fruit because fruit are rejected from market on the basis of presence or absence of disease, not on its severity.

The disease incidence variables we have used are recorded on specified types of shoot. For black spot we record, 1) the number of leaves with lesions as a percentage of the total number of leaves per shoot, and 2) the percentage of fruit with lesions. For powdery mildew we record, 1) the number of leaves with lesions as a percentage of the total number of leaves per shoot, 2) percentage of shoots with mildew infection of the growing apex and 3) percentage of fruit with powdery mildew-induced russeting.

Because only young leaf tissue is susceptible to black spot and powdery mildew infection, the rate of new leaf emergence is particularly important in relation to disease development. Disease assessments should therefore focus on extension shoots of type 1) and 2) and possibly 4), where new leaves provide potential for continued infection throughout the season.

Disease can decrease during the season. For fruit, this is caused by natural abscission and by chemical or hand thinning. Damaged fruit is often selectively hand thinned, and disease assessments before and after thinning may be required to take account of this. Disease incidence may decrease as a result of emergence of new leaves (Jeger 1981) and because of leaf abscission. Recording numbers of leaves per shoot allows the "diluting" effect of new leaf emergence on disease to be determined, and the effects of leaf drop can be determined by counting the number of leaves present in relation to the number of leaf scars on the stem.

Black spot and powdery mildew symptoms are generally distinctive, however it may be necessary to check microscopically for presence of conidia of *V. inaequalis* or *P. leucotricha* in the types of necrotic lesions which are being observed. Powdery mildew colonies become brown as they age and can be confused with other types of leaf necrosis. Black spot lesions can also be confused with other leaf necroses late in the season. Powdery mildew may be difficult to recognize on the underside of leaves in cultivars where leaves have dense white hairs on their lower surface.

Different types of black spot lesions can be recognized, eg. actively sporulating versus inactive, or discrete versus "sheet scab" lesions. Abaxial leaf surfaces can be assessed separately from adaxial surfaces. Only the information that is relevant to pre-planned objectives should be recorded, as each additional level of detail increases assessment costs and complicates data interpretation.

Sampling procedures

Assessments are carried out non-destructively on 10 randomly selected shoots of specified type per tree, on each of 4-6 trees per experimental plot or orchard block. For fruit assessment, the number of fruit per cluster can be recorded (Table 1), but a simple incidence assessment for black spot and for russet (Faust and Shear 1972) on 50-100 fruit per tree is often all that is required. These sample sizes are suitable where disease incidence is greater than about 1%. If incidence is lower, larger numbers of leaves and fruit must be sampled by increasing both the number of trees sampled and the number of shoots per tree. When repeated sampling is carried out, the same trees are sampled at each date with shoots and fruit selected at random each time. Greater precision can be obtained by tagging shoots and assessing the same shoots at each date, but there is a high time cost in placing tags, particularly when disease incidence is low.

Because it is difficult to see the whole fruit surface, non-destructive assessments tend to underestimate fruit damage and an assessment of a picked sample (Croxall *et al.* 1952b) may be required for better accuracy. Apical mildew infection causes deformed leaves and precludes any other measurements being taken on affected shoots. If the incidence of apical mildew is greater than about 5%, additional shoots should be sampled to maintain the sample size for other variables.

The timing of assessments depends on the objective of the work, but for assessment of disease where fungicide chemicals or cultivars with differing susceptibility are compared, at least two samples are required, one in early December, and one immediately before the harvest of each cultivar. For disease management, a single assessment in early December allows black spot incidence at the end of the ascospore release season to be used to plan the fungicide programme for the rest of the season. The data collected using these methods have several spatial sampling strata (shoot, tree, plot or orchard block) and are suitable for analyses of spatial variability or pattern of disease occurrence.

This paper describes methods for the assessment of apple diseases which have been refined over 7 years to allow efficient data collection. They have been used in field trials comparing the efficacy of fungicides, susceptibility of cultivars and the timing of fungicides in relation to weather information for disease management. Future uses will include investigation of interactions between the host plant and disease, and the collection of data for on-orchard computer data bases for practical disease management systems (Laurenson *et al.* 1994).

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