



**BIOSECURITY SA  
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# Compendium of branched broomrape research

## Section 13. Assessing eradication

A COMPILATION OF RESEARCH REPORTS FROM THE  
BRANCHED BROOMRAPE ERADICATION PROGRAM SOUTH  
AUSTRALIA

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**PREMIUM**  
FOOD AND WINE FROM OUR  
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ENVIRONMENT



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See also the following publications:

Panetta, F.D. and Lawes, R. (2005). Evaluation of weed eradication programs: the delimitation of extent. *Diversity and Distributions* 11, 435-442.

Panetta, F.D. and Lawes, R. (2007). Evaluation of the Australian branched broomrape (*Orobanche ramosa*) eradication program. *Weed Science* 55, 644-651.

Panetta, F.D., Cacho, O., Hester, S., Sims-Chilton, N. and Brooks, S. (2011). Estimating and influencing the duration of weed eradication programmes. *Journal of Applied Ecology* 48, 980-988.

Prider J., Correll R., Warren P. (2013) A model for risk-based assessment of *Phelipanche mutellii* eradication in fields. *Weed Research* 52, 526-534.

# 1. Method for assessing seed viability for determining status of infested paddocks

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*April 2003*

## Background

The Branched Broomrape Program is working on the strategy for paddocks (and eventually properties) to be released from quarantine. Release from quarantine will rely on paddocks being kept free of seeding branched broomrape plants for a specified number of years.

Paddocks must be kept free of branched broomrape plants for seven consecutive years to achieve 'Provisional' status. After a further five consecutive years without branched broomrape plants setting seed, paddocks will achieve 'Eligible' status. Once all paddocks on the property are eligible for release, the Property becomes eligible for release from quarantine.

If branched broomrape plants emerge, farmers have a small window of opportunity in which plants can be treated to prevent seed set. If they are successful in doing this, their paddock retains Provisional status on the way towards release from quarantine. If they are unsuccessful, paddocks return back to year 1 on the timeline for release.

## Proposal

The Broomrape Program has a means of testing whether or not seed collected in the field is viable. Seed is subjected to GR24 solution which encourages germination. Germinating tubercules can be viewed under a microscope two weeks after the application of GR24. This is a measure of how many seeds are viable.

If branched broomrape plants emerge and are treated by the landholder, seed needs to be tested to determine whether or not the treatment was successful. A sample of seed can be collected and subjected to GR24 to determine viability.

The Broomrape Program needs a suitable methodology for collecting seed from infested paddocks to test whether or not viable seed was set. Infestations will vary in size from 1 plant up to many dozens of plants. The density of plants will vary from one plant per paddock to many hundreds of plants per hectare. When infestations are found, and before they are treated, they are accurately mapped using a Differential Global Positioning System.

## Our Requirement

That a statistical test be developed to determine whether or not viable seed has been produced in paddocks infested with branched broomrape. The test should include a suggested survey design for seed sampling transects through an infestation that has already been well defined during earlier surveys. The test should also include the number of plants which need to be sampled per linear metre of infestation to collect meaningful data on the viability of seed in the paddock.

## Statistical Response

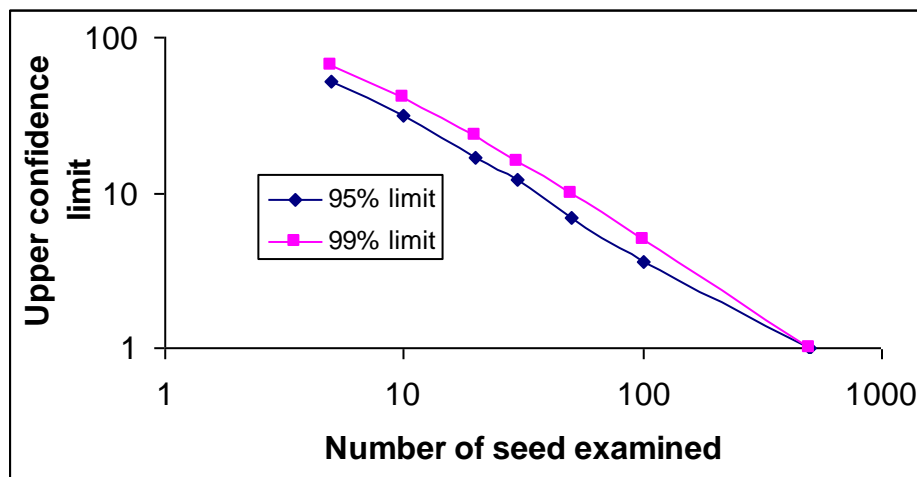
### Number of seeds

The number of seeds to be tested (assuming that the test is very effective) is given in Table 1. The same data are illustrated in

Figure 1.

**Table 1. Upper confidence limits of % seed that are viable when no positives are recorded**

Number of seeds examined	95% limit	99% limit
5	52	65
10	31	41
20	17	23
30	12	16
50	7	10
100	4	5
500	1	1



**Figure 1. Upper confidence limits of % seed that are viable when no positives are recorded**

The implication of those data is that about 100 seeds need to be tested to establish that the germination rate is no more than 5%. If a more rigorous standard is required, more seeds need to be considered. For example 500 seeds are required to be sure that the germination rate is no more than 1%.

Scenario 1 – High density infection (100 plants per hectare, with each plant producing 10,000 seeds that are scattered across the entire area).

In this scenario, the average seed density is 100 per square meter. If we assume that sampling is to about 70 mm and the bulk density is about 1.4 kg/L, 100 kg of soil would need to be examined to obtain sufficient seed to obtain 100 seeds.

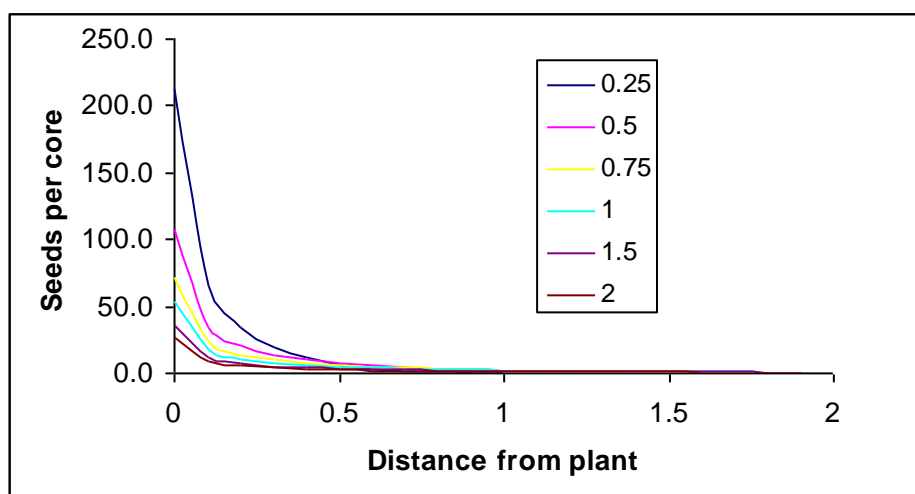
Scenario 2 – Moderate density infection (1 plant per hectare, with each plant producing 10,000 seeds that are scattered across the entire area).

Using similar assumptions to Scenario 1, 10 tonnes of soil would need to be sampled completely to obtain the required 100 seeds.

Scenario 3 – Low density (1 plant per hectare, with each plant producing 10,000 seeds 10% of the seeds remaining and these are scattered across the entire area).

Using similar assumptions to scenario 1, 10 tonnes of soil might need to be processed to locate a single seed. With such mass processing, the efficiency of finding seeds would be less than 100% implying that even more soil has to be processed.

Scenario 4 – Isolated plants, seeds scattered in a random direction, 50% of seed within say 1 m of the plant, 10,000 seeds initially reduced to 1000.



**Figure 2. Predicted number of seeds in cores taken near a single broomrape plant**

In this scenario we may also vary the radius that includes half the seeds. We further assume that the distance a seed is from the plant is normally distributed. Initially we assume that there is no directional preference of the seeds, but this assumption is not critical.

We assume that the cores 100 mm diameter taken to a depth that includes all the seeds.

The above assumptions lead to the data shown in

Figure 2 and Table 1.

### Alternative variable to measure

An alternative approach that may be more profitable is to consider the number of viable seeds per unit area. The same coring approach would be used for this as for the percent of seeds that is viable.

### Sampling seeds from plants

#### Estimation of viability of seeds from plants

Under some circumstances it will be desirable to estimate the proportion of viable seeds by sampling plants.

The following protocol should be used in such circumstances.

Sampling of seeds directly from plants is a viable option. The sample of seeds taken should be representative of the seeds in the field.

Sampling of plants should be avoided if possible – rather all the plants should be gathered. This should be feasible if there are no more than 50 plants in the patch.

**Table 2 Expected number of seeds in 100 mm cores taken at varying distances from a broomrape plant, where half the seeds stay within a given radius, assuming that 1000 seeds have survived to the time of sampling**

Radius	0.25	0.5	0.75	1	1.5	2
Distance						
0	212.7	107.3	71.7	53.8	35.9	26.9
0.1	65.9	35.1	23.7	17.8	11.9	9.0
0.2	34.2	20.3	14.0	10.6	7.1	5.4
0.3	19.7	13.7	9.8	7.5	5.1	3.8
0.4	11.5	9.9	7.3	5.7	3.9	3.0
0.5	6.5	7.4	5.8	4.6	3.2	2.4
0.6	3.6	5.6	4.7	3.8	2.6	2.0
0.7	1.9	4.3	3.8	3.2	2.3	1.7
0.8	0.9	3.3	3.2	2.7	2.0	1.5
0.9	0.4	2.5	2.6	2.3	1.7	1.3
1	0.2	1.9	2.2	2.0	1.5	1.2
1.1	0.1	1.4	1.8	1.7	1.4	1.1
1.2	0.0	1.0	1.5	1.5	1.2	1.0
1.3	0.0	0.8	1.3	1.3	1.1	0.9
1.4	0.0	0.5	1.1	1.2	1.0	0.8
1.5	0.0	0.4	0.9	1.0	0.9	0.8
1.6	0.0	0.3	0.7	0.9	0.8	0.7
1.7	0.0	0.2	0.6	0.8	0.8	0.6
1.8	0.0	0.1	0.5	0.7	0.7	0.6

Where there are more than 50 plants in a patch a sampling scheme will be used. The basic technique to be used will be a belt sample. The procedure will be as follows

1. Ascertain the area  $A$  and width  $W$  (East-West) of the patch from previous GIS data;
2. Collect all plants in a belt transect that runs from North-South through the patch and count the number  $n$  in the transect;
3. Estimate the number of transects as  $T = 100/n$
4. Estimate the distance apart of the transects as  $D = W/T$

Walk and collect all the plants in each belt transect which will be running North-South, with centre of each transect a distance  $D$  from the centre of the previous transect. Transects will be walk both to the East and the West of the initial central North-South transect.

The following should be noted in conjunction with the above procedure –

1. The scheme is targeted at obtaining between 50 and 100 plants. If the patch is circular about 75 (or more exactly  $100\pi/4$ ) would be expected in the sample.
2. The scheme envisages a belt with of about 2 m, but this is distance is not critical so long as the belt width is kept more or less constant across the sampling. The transect width  $w$  should be chosen to be as wide as feasible so that every broomrape plant in the transect will be observed.
3. An estimate of the number of plants  $N$  in the patch can be obtained from the formula

$$N = \frac{mW}{wT}$$

where  $m$  is the total number of observed plants in all the transects, and  $w$  is the transect width. An approximate standard error of that estimate is

$$SE = \left( \sqrt{1 - \frac{wT}{W}} \right) \frac{W}{wT} \sqrt{m}$$

4. If the plants are of similar size, an equal number of seeds should be taken from each plant. If there are large differences between the plants, more seeds should be taken from the larger plants (ideally the number of seeds taken from each plant should be proportional to the size of the plant).
5. The sampling assumes that all the seeds have been retained in the plants. If this assumption is not valid it is implicitly assumed that proportion of viable seeds is the same for shed and unshed seeds.



## 2. Risk of broomrape recurrence in paddocks

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*June 2010*

### Summary

Release of paddocks from quarantine requires balancing the risk of recurrence of branched broomrape against the benefit of release from quarantine. This report uses discovery survey data to measure the interval between years of branched broomrape detections in a paddock.

Using ten years of survey data, the proportion of surveyed paddocks with increasing intervals of no branched broomrape detection has been calculated. Data from the last five years showed that the longer the interval since detections the lower the risk of broomrape recurrence. This relationship held for different survey years even where there had been variations in the overall levels of detection.

Predictive modelling based on the last three years of survey data, indicates that after a branched broomrape-free interval of 12 years in a paddock, there is less than a 1% probability of broomrape recurrence.

### Introduction

Branched broomrape is a noxious weed that is currently confined to a quarantine area in South Australia. The management of this weed is aimed at eradication. Two criteria for the success of the program are the number of paddocks where branched broomrape is observed for the first time (new discoveries) and the number of paddocks where branched broomrape has been observed in a paddock subsequent to a discovery but not necessarily from a consecutive survey (returns). Results from annual surveys have been encouraging with a decreasing number of new discoveries and a decreasing return rate.

Farmers within the quarantine area have restrictions on the movement of product off their properties. There is therefore pressure to release paddocks from quarantine. It is practically impossible to guarantee that a paddock is in fact free of branched broomrape. Release from quarantine thus becomes a matter of balancing the risk of a recurrence of branched broomrape against the benefit of release from quarantine. This report explores the risk of a recurrence.

It is reasonable to assume that the risk of a recurrence would decrease with an increasing interval of no branched broomrape detections in that paddock. Data are available to quantify this assertion. There is however confounding of this apparent effect with a general decrease in the number of detections (both new discoveries and recurrences). This report has therefore isolated these effects by performing separate analyses for each target year.

### Data

Analyses were based on the discoveries data base that is maintained by Mr Nick Secombe. This database includes the results of annual discovery surveys within paddocks within the Quarantine Area and selected adjoining paddocks between 1999 and 2009. The analysis is based on the recorded presence or absence of branched broomrape in a paddock each year. Not all paddocks were surveyed in all years as the Quarantine Area has not remained static.

## Statistical Methods

The number of branched broomrape free years was defined as the difference between an initial observation year and the year of the nominated observation. The earliest observations were in 1999 and the latest were in 2009 making the potential number of free years as 10. For simplicity we denote the number of free years as the gap.

The observed rate of return after a number of free years was calculated as the observed number of eligible positive paddocks divided by the total number of eligible paddocks. For a paddock to be eligible it must meet the following criteria:

- The paddock must have been positive in the initial observation year;
- It must have been surveyed in the nominated year; and
- There must be no occurrence of branched broomrape during the gap.

A paddock was still considered eligible if a survey had been missed in the gap period, It was also considered eligible if there had been a return infestation prior to the initial observation.

The proportion of paddocks for each gap length were calculated and plotted against the gap length. This was repeated for each observation year, starting with year 2000 and repeating up to 2009.

The proportion of infested paddocks was modelled using a generalised linear model using binomial errors and a logistic link. The corresponding fitted proportions were plotted on the same graph, noting that

$$p = (\exp (l))/(1 + \exp (l))$$

where  $l$  is the predicted value on the logistic scale. The predicted value of  $l$  for 10,12, 15 and 20 years was calculated from this model.

## Results

Table 3 shows the eligible paddocks for each of the survey years. Year 1 includes the paddocks that were observed with branched broomrape the previous year, that were surveyed in the nominated year. Year 2 includes those paddocks that have had a branched broomrape free gap of two years, year 3 paddocks have been broomrape free for three years etc.. The criteria for a paddock being eligible are given in the Statistical Methods section of this report.

**Table 3. Number of eligible paddocks that could potentially have returned an infestation**

Year	Time since last observed									
	1	2	3	4	5	6	7	8	9	10
1999										
2000	82									
2001	239	54								
2002	263	167	35							
2003	99	223	150	30						
2004	365	54	122	112	23					
2005	93	306	51	113	107	23				
2006	158	58	244	48	105	93	20			
2007	195	114	41	207	47	89	87	16		
2008	131	148	96	34	183	44	85	86	14	
2009	94	102	130	89	33	177	44	82	84	14

**Error! Not a valid bookmark self-reference.** shows the number of eligible paddocks where branched broomrape was detected during each of the nominated survey years. Up until 2005 the number of paddocks with returning branched broomrape has been variable but since that time there has been a decrease in the recurrence rate. In the last three years, recurring paddocks are predominantly those where branched broomrape has been recorded in the previous four years.

**Table 4. Number of eligible paddocks that returned an infestation**

Year	Time since last observed										Total paddocks returning	
	1	2	3	4	5	6	7	8	9	10		
1999												0
2000	28											28
2001	72	19										91
2002	40	17	5									62
2003	44	101	38	7								190
2004	62	4	9	5	0							80
2005	35	63	4	8	16	3						129
2006	44	16	40	1	16	6	4					127
2007	47	18	8	24	2	4	1	2				106
2008	29	18	7	1	6	0	3	2	0			66
2009	16	11	6	4	0	4	0	1	1	0		43

**As the intervals between detection of branched broomrape in a paddock increase, the risk of branched broomrape recurrence decreases (**

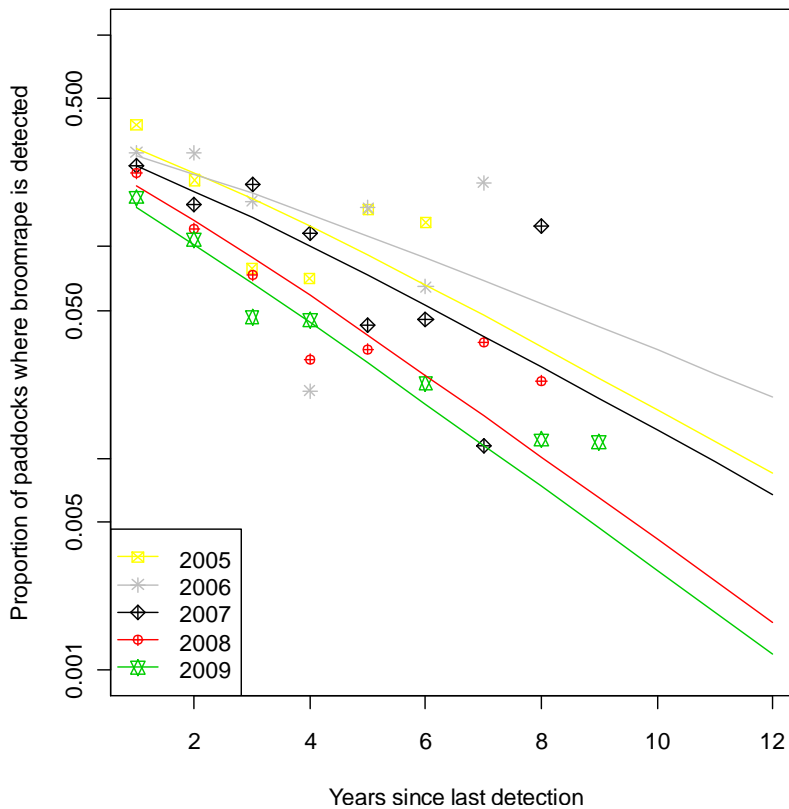
Figure 3). Although there has been a reduction in discoveries over this time, as shown by the different curves for each survey year in Figure 1, the risk of recurrence has maintained a negative decline. Note

that the points for the 10 year gap in the 2009 series was not plotted because it was zero; the 9 year gap in the 2008 series was also zero. The rate of decline, as indicated by the slope parameter in Table 5, was higher in the 2008 and 2009 data series than in the preceding series but as can be seen from standard errors of the slopes the change was not statistically significant therefore.

Predictions calculated from the recurrence data of the last three years show that the risk of branched broomrape recurring in a paddock after an absence of 12 years is less than 1 % (Table 6).

**Table 5. Details of logistic model as shown in Figure 1 for years 2005 to 2009. The ‘X’ value has been centred on a gap of 12 years so the intercept represents the predictor at 12 years**

	Intercept		Slope	
	Estimate	Standard error	Estimate	Standard error
2005	-5.112	0.846	-0.351	0.080
2006	-4.191	0.630	-0.266	0.061
2007	-5.337	0.634	-0.349	0.060
2008	-6.834	0.821	-0.451	0.077
2009	-7.187	0.919	-0.456	0.088



**Figure 3. Affect of increased years since last detection on the proportion of paddocks returning positive**

**Table 6. Percentages of paddocks where branched broomrape will be detected following a given branched broomrape-free interval**

Survey year	Branched broomrape free interval (years)			
	10	12	15	20
2005	1.70%	0.85%	0.298%	0.052%
2006	3.25%	1.94%	0.881%	0.234%
2007	1.35%	0.68%	0.239%	0.042%
2008	0.41%	0.17%	0.044%	0.005%
2009	0.30%	0.12%	0.030%	0.003%

## Discussion

There has been a steady decline in the rate of recurrences while the management practices have remained similar. If, as theory indicates, the number of attachments is the product of the seed density and the host density, this strongly implies that the seed density is declining.

The lowering with time of the predicted probability of a recurrence following a gap of 12 years is also encouraging. With only 14 paddocks eligible for release in 2012, the probability of a recurrence would be approximately 1% across all released paddocks. In subsequent years with larger numbers of paddocks due for release – this larger number of paddocks will have to be monitored carefully as the probability of a recurrence will be approaching 10% across all released paddocks as a result of the summation of the individual probabilities of recurrence for each paddock. Further data are required to determine whether the downward trend in the estimate of the probability of recurrence continues.