Dispersal biology of *Orobanche ramosa* in South Australia

Master of Science Thesis

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Chapter 3. Natural wind dispersal of Orobanche ramosa

Introduction

O. ramosa is an annual, holoparasitic weed that predominantly flowers during spring time in South Australia. Seeds are approximately 0.3 mm by 0.2 mm in size, weigh 3 to 6 μg (Parker and Riches 1993), are ellipsoid to ovoid in shape (Plaza et al. 2004), and the seed coat has a raised ridged pattern with a series of reticulated, polygon cells (Parker and Riches 1993). Some *Orobanche* species can produce up to 10 000 seeds per capsule (Molau 1995), and these are very long-lived; *O. ramosa* seed has been reported as surviving for up to 13 years in soil (Parker and Riches 1993).

The small 'dust' like seeds of *Orobanche* species suggest that wind may be a dispersal mechanism due to the large surface area to weight ratio (Howe and Smallwood 1982; Hughes et al. 1994), although elaborate wings and plumes are absent. Kuijt (1969) suggests that while the concave cavities on the surface of *Orobanche* species seeds (shown in Figure 3.1, Plaza et al. 2004) may assist seed to lift up in wind, the small size might negate these as aids for wind dispersal.

One of the few papers investigating wind dispersal of small seeded parasitic species is by Berner et al. (1994) who found that *Striga hermonthica* (Del.) seeds dispersed up to 12 m horizontally and 2 m vertically by wind in fields of maize and sorghum in Nigeria. However they concluded that wind was a relatively unimportant vector for *S. hermonthica* compared with human-induced movement through agricultural practices.

In seed dispersal theory two stages of seed dispersal are recognised. Phase I (Chambers and MacMahon 1994) or local dispersal (Cousens and Mortimer 1995) is the initial separation of the seed from the parent plant and its settling onto a surface. Phase II (Chambers and MacMahon 1994) or geographic dispersal (Cousens and Mortimer 1995) is the subsequent movement of the seed from that first surface, to another location. While it is difficult to completely separate the two stages in a field experiment, this chapter attempts to document Phase I or primary dispersal of *O. ramosa* seed from the adult plant.

This chapter investigates primary wind dispersal of *O. ramosa* seeds under field conditions. The aims were:

- To determine if the direction of *O. ramosa* seed dispersal is correlated with wind direction, under field conditions in the quarantine zone in South Australia; and
- ii. To determine how far *O. ramosa* seeds can be blown by wind in primary dispersal under field conditions.

Methods

Field site

The study site was located 71 km east of Adelaide, near Mannum, South Australia, (34.91°S, 139.40°E), on a farming property within the South Australian Branched Broomrape Program quarantine zone.

The field survey was conducted from 26 November to 2 December 2004.

Seed sources

Two plots, 25 x 7 m, free of emerged *O. ramosa* plants were selected; one a standing wheat crop, the other a sparse *Medicago* sp. pasture. Mature, seed-bearing *O. ramosa* plants were removed by hand from elsewhere on the site where they were parasitising *Hedypnois rhagadioloides*, and arranged into groups of stalks with 30 mature seed capsules. Remnant flower petals were removed from the seed capsules where present, along with snails and other foreign material. Seed presence was tested by removing a single capsule from each stalk and breaking that capsule to expose the seeds. If seeds were present in the tested capsule, that whole stalk was used. If there were no seeds in the tested capsule, the stalk was discarded. Groups of stalks with 30 seed-bearing pods were attached with sticky tape to a wire pin, which was stuck into the ground so that seed pods were > 0.1 m above the soil surface.

Two sets of stalks were installed into each plot, approximately 5 m apart: two in pasture and two in cereal, giving two replicates in space for pasture and cereal. Traps were redeployed every 24 hours for five consecutive days (starting 26 November

2004), with new seed sources installed each day, giving five replicates in time; in total n = 10 for pasture and n = 10 for cereal.

Seed traps

A number of seed trap designs were trialled before the survey began, see Appendix 2 for details.

One seed trap consisted of 19 abutting glass microscope slides 76 x 25 mm (Livingstone International Pty Ltd, Rosebery, NSW) placed on an aluminium rack 150 x 3 x 1 cm. A single continuous strip of double-sided tape 150 x 1.8 cm wide (Sellotape Regd. Dalton Packaging Pty Ltd Bankstown, NSW) was placed down the centre of the slides, secured at each end by sticking the tape to the aluminium. Eight traps were placed on the ground around a single plant in eight cardinal directions: north, north-east, east, south-east, south, south-west, west, and north-west (Figures 3.2 and 3.3)

Traps were placed into position around the seed source, and the backing paper from the double-sided tape was removed. Traps were retrieved and replaced every 24-h. On retrieval, traps were lifted from the ground and clean, single-sided sticky tape was placed over the top of the double-sided tape to seal the sticky surface with the seeds held in place. Traps were labelled and bagged for counting in the laboratory.

Traps were analysed under a stereo dissecting microscope. Seeds on each segment of the traps (slides) were counted and recorded. It was assumed that trapped seeds came from the nearest experimental seed source.

Wind

Temperature, wind speed and wind direction were measured and recorded. Two anemometers were set up on site. The site-level anemometer was secured to a metal pole and positioned 1.7 m off the ground in the middle of the experimental area (Figure 3.4). The ground-level anemometer measured wind speed at a height of 0.16 m (Figure 3.5). The unit was movable and could be placed adjacent to the seed traps and translocated 'plants' to accurately measure the wind speeds being experienced by the seeds. The anemometer was secured within a portable open 'cage' that prevented any interference by plant material. Each anemometer was connected to a data logger.

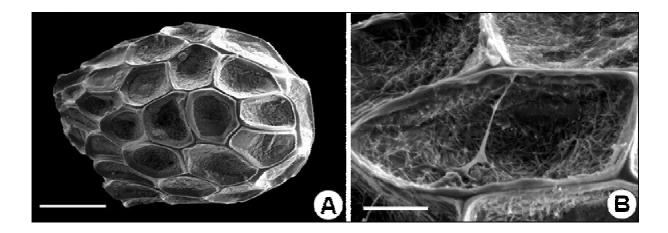


Figure 3.1. Scanning electron micrographs of *Orobanche* species seed. A. *O. latisquama*, the seed size and shape is similar to *O. ramosa*, scale bar = $100 \,\mu\text{m}$. B. Close-up view of *O. ramosa* seed coat, showing reticulated, polygon cells separated by ridges, scale bar = $20 \,\mu\text{m}$. Both images copied from Plaza et al. (2004).

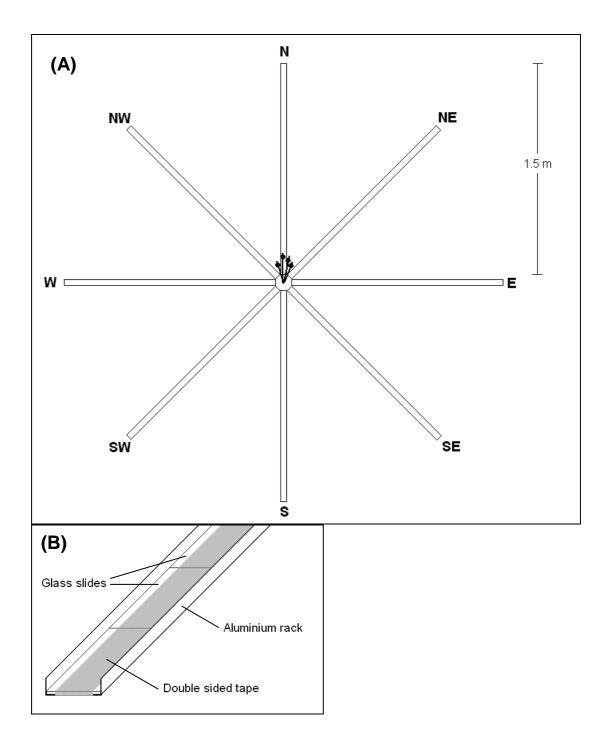


Figure 3.2. Seed trap design. (A) Eight aluminium racks were placed in eight cardinal directions around a transplanted seed source. (B) Seed traps consisted of 19 abutting glass microscope slides placed on each aluminium rack 1.5 m long. A single continuous strip of double-sided tape was placed down the centre of the slides.

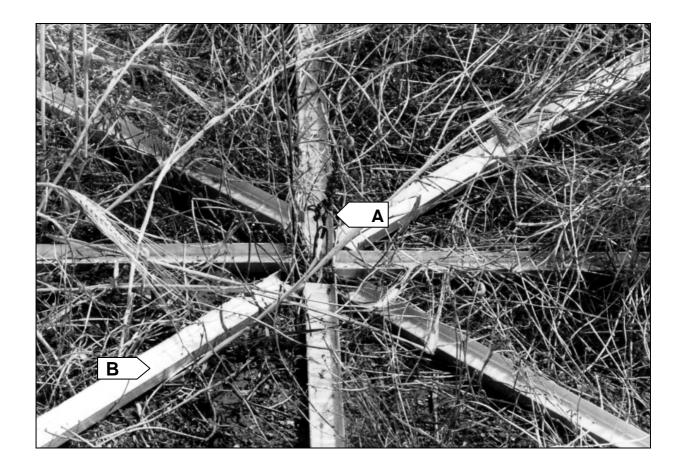


Figure 3.3. Seed traps used to survey natural wind dispersal from mature seed-bearing *O. ramosa* plant. (A) Dark, mature *O. ramosa* plant, surrounded by eight seed traps (B) radiating in eight cardinal directions.

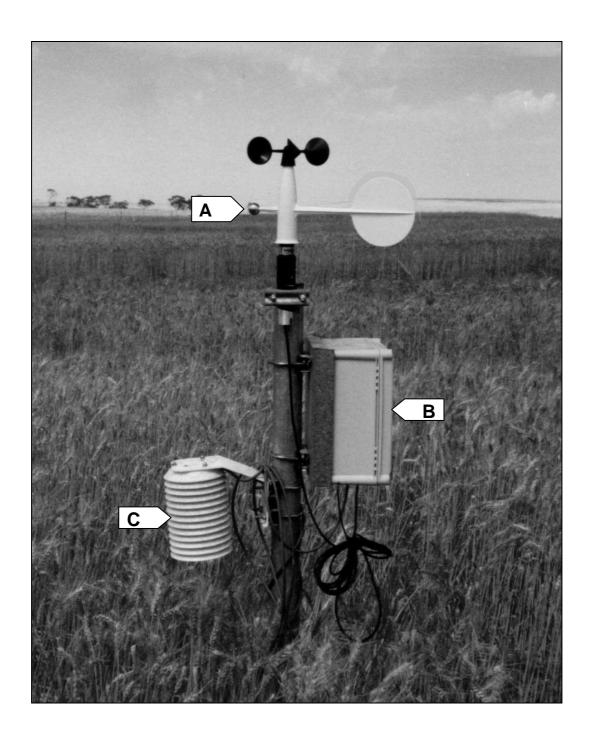


Figure 3.4. Field set-up for measuring and recording wind speed, wind direction and air temperature. (A) Anemometer and wind vane, (B) data logger, and (C) relative humidity sensor.



Figure 3.5. Field set-up for measuring wind speed at ground-level. (A) Anemometer in 'cage' to protect cups from interference by vegetation, and (B) the data logger to collect the information.

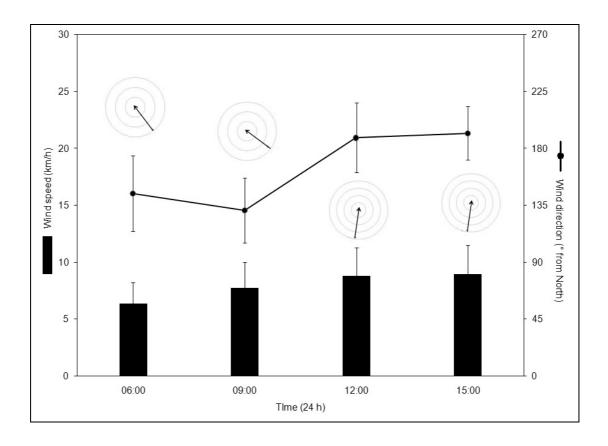


Figure 3.6. Wind speed and direction for Murray Bridge, at 0600, 0900, 1200 and 1500 h, 26 November 2004 to 8 December 2004 inclusive. Data are means \pm SE, n = 12. Data provided by Bureau of Meteorology. Arrows in circles indicate mean wind direction (in ° from North, 0°) for each time period.

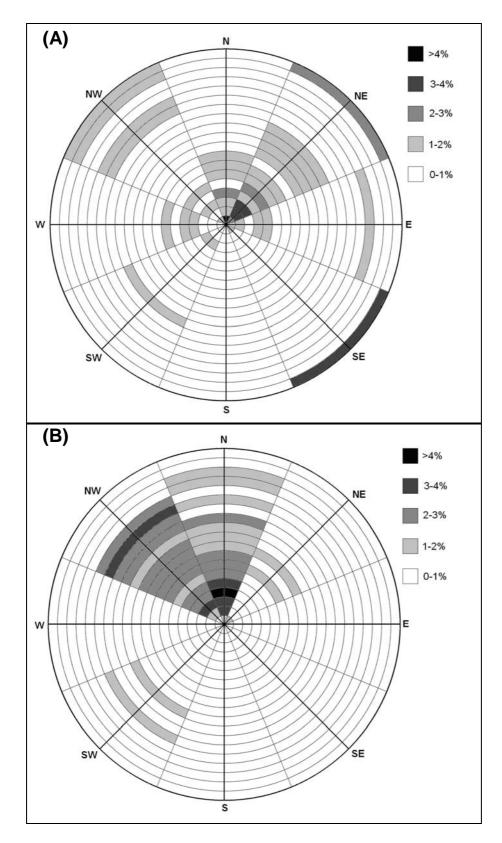


Figure 3.7. *O. ramosa* seeds trapped from a point source in each of the eight cardinal directions per 24 h under field conditions. Data from all replicates are pooled for each treatment (A) cereal n = 9, and (B) pasture n = 10.

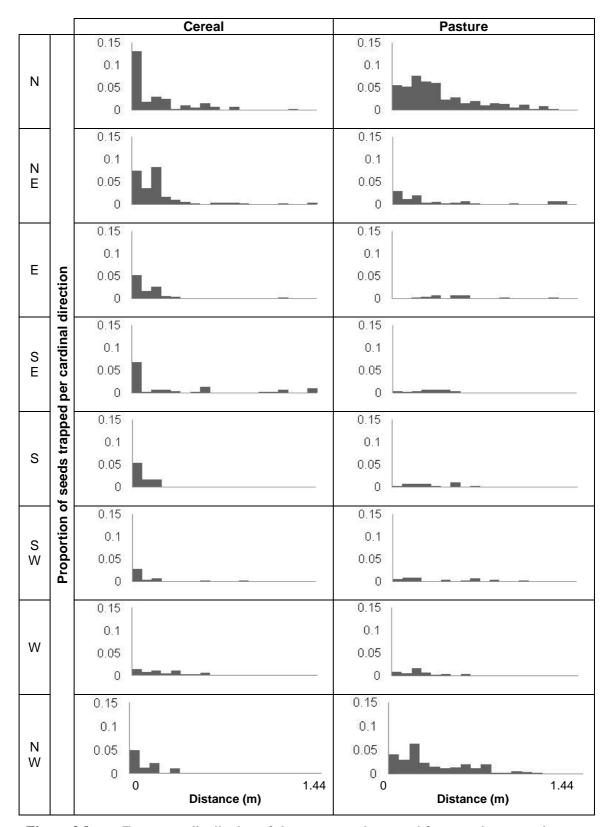


Figure 3.8. Frequency distribution of *O. ramosa* seeds trapped from a point source in cereal (left column) and pasture (right column). Data are proportion of seeds trapped per cardinal direction; histograms indicate mean number of seeds in each cardinal direction, n = 9 for cereal and n = 10 for pasture.

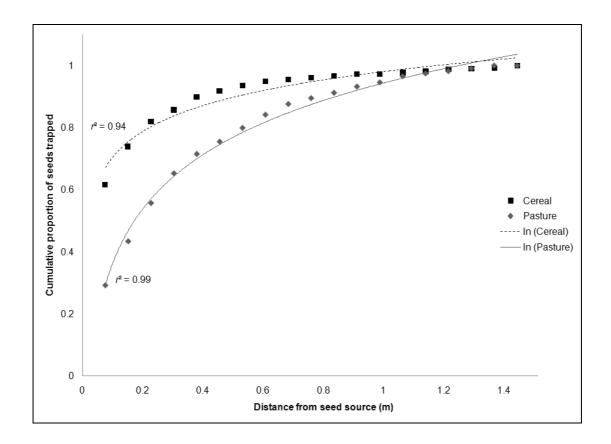


Figure 3.9. Cumulative seed dispersal distance for *O. ramosa* in cereal and pasture (cereal n = 9, pasture n = 10). Data are means of proportion (per replicate) of seeds pooled for all cardinal directions, with fitted natural logarithmic trend lines, r^2 values as shown on the graph. Standard error bars are not shown as they were very large.

Unfortunately, the data from both data loggers were lost, and so wind data were sourced from the nearest weather station of the Australian Bureau of Meteorology. Data for wind speed and direction were obtained for the Murray Bridge site (approximately 27 km in a direct line from the study site, 35.12°S, 139.26°E, elevation 33 m) at four times during the day (0600, 0900, 1200 and 1500 h) over the course of the experiment.

Results

Wind

The Bureau of Meteorology data showed that wind was blowing from the south-east at velocities ranging from 6 to 8 km h⁻¹ in the mornings (0600 and 0900 h) and from the south at velocities of 8 to 9 km h⁻¹ in the afternoons (1200 and 1500 h) during the survey period (Figure 3.6).

Seed survey

One of the cereal replicates was ruined by hares digging near the traps throwing sand over the traps, and so the numbers of seeds trapped was not able to be quantified for that replicate. The final number of replicates that yielded useable data totalled 19, 9 for cereal and 10 for pasture. Of those 19 replicates, the mean number of seeds trapped per replicate was 99 (SE = 28.8, min = 1, max = 472).

Primary seed dispersal, as indicated by the percent of seeds trapped in the eight cardinal directions, was predominately in the N-NW direction for pasture, and variable for cereal (Figure 3.7).

Over the trapping distance (1.44 m from seed source) the majority of seeds were trapped close to the source, regardless of direction (Figure 3.8). The number of seeds trapped decreased with increasing distance from the seed source (Figures 3.8 and 3.9). In cereal, 90% of *O. ramosa* seeds were trapped within 0.38 m of the source, whereas 90% of *O. ramosa* seeds were trapped within 0.76 m in pasture (Figure 3.9).

Discussion

A correlation was shown between wind direction and the direction of *O. ramosa* seed trapped in pasture. The pattern of seed detection was less clear in cereal, but there appeared to be something of a northerly trend. Over the trapping period, the prevailing wind was from the south-east and south, while seeds were trapped

predominantly in the north and north-westerly direction. Similarly, Bullock and Clarke (2000) showed that seeds typically dispersed in the direction of the 'cumulative wind speed' (a measure of wind velocity and proportion of time wind was blowing in a particular direction).

Mesa-García et al. (1986) hypothesised that the direction of crop rows in a field (in their case, broadbeans) may influence direction of wind dispersed *O. crenata* seeds, but that this would be secondary to the influence of mechanical tillage and harvesting. In this experiment, our cereal rows were oriented approximately north-south. If the wind acting on the *O. ramosa* seeds were being funnelled by the cereal, we would expect to see the seeds in cereal oriented to the north and north-west directions, whereas the results indicate the seeds in cereal were trapped in many different directions.

As described in the methods section, the initial methodology allowed for site level and ground level wind data to be collected during the trapping period via two anemometers. However the data was lost before analysis could be carried out. The wind data sourced from the nearest Bureau of Meteorology weather station, Murray Bridge, was presented as daily data for each of the trapping days, with wind speed (in km h⁻¹) and direction (compass points) collected at 0600, 0900, 1200 and 1500 h. This 'snap shot' style of data capture did not show stochastic wind events that may be important dispersal vectors (Dauer et al. 2007); indeed Salisbury (1942) found that wind gusts were important in facilitating the release of poppy species (as cited in Cousens et al. 2008, p. 22). Additionally, the weather station was located approximately 27 km from the study site, and may not accurately reflect the microclimate of the study area. An additional limitation in the methodology was the use of wire pins to secure the translocated stalks into the centre of the seed traps. The presence of the wire may have reduced the ability of the plant to bend and sway in gusty wind, and thus may have reduced the ability of the seeds to disperse naturally. There was a sharp decline in numbers of seeds trapped with increasing distance from the source, approaching a negative exponential decline in seed number with distance. This result mirrors the wind dispersal pattern for *Striga* species found by Berner et al. (1994) and is similar to patterns found in other seed dispersal studies (Bullock and Clarke 2000).

While the majority of the seeds trapped were caught close to the seed source, seeds were also trapped up to 1.44 m from the seed source. Thus, *O. ramosa* seeds are able to be dispersed at least that far under low wind speeds (mean wind velocity 8 km h⁻¹).

The methodology used here did not attempt to gain a true estimate of the long distance dispersal tail for wind dispersed *O. ramosa* seeds. Should future experimenters wish to document the tail of the distribution curve, they should follow the advice of Bullock and Clarke (2000), whose methodology, though laborious, used increasing numbers of traps placed with increasing distance from the seed source to maintain a large sampling intensity for small seeded plants.

On average more *O. ramosa* seeds were trapped closer to the base of the plant in cereal plots compared with pasture. This may be due to the taller cereal ground cover reducing the wind velocity acting on the dispersing seeds, thus reducing the distance of dispersal in cereal compared with pasture.

This survey of *O. ramosa* seed dispersal was intended to document Phase I or primary dispersal only. It is assumed that the seed trap design trapped seeds in the process of primary dispersal, but this assumption has not been tested. While this survey showed that *O. ramosa* seeds are readily dispersed at least short distances by low wind, it does not rule out other vectors acting in parallel. Indeed, agricultural practices that move soil (such as tillage and harvesting) may be a more important vector for *Orobanche* species seed in a cropped field situation (Mesa-García et al. 1986).

This investigation, designed to survey the primary dispersal of *O. ramosa* seeds in the field with wind conditions typical of those experienced by mature seed-bearing plants, has allowed basic inferences to be made about wind dispersal in this species. The following chapter describes the use of a portable wind tunnel to investigate the dispersal of *O. ramosa* seeds under different wind speeds and surface types in the field.