



Testing the consistency of spatio-temporal patterns of flight activity in the stored grain beetles *Tribolium castaneum* (Herbst) and *Rhyzopertha dominica* (F.)



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ABSTRACT

Many studies have been published on the flight activity of stored grain beetles in temperate regions, but few have focussed on tropical and sub-tropical regions. The spatio-temporal patterns of flight activity of *Tribolium castaneum* (Herbst) and *Rhyzopertha dominica* (F.) were quantified in a grain growing district on the Tropic of Capricorn in central Queensland, Australia. Nine traps baited with aggregation pheromone lures were monitored at 2-wk intervals (fortnightly) for 1 year along a 28.4 km linear transect that included sites at bulk grain depots and sites away from stored grain. Beetles of both species were trapped every fortnight during the study. The spatio-temporal patterns of flight activity differed greatly across the two species, as predicted from studies elsewhere. *Rhyzopertha dominica* was widespread across the landscape, as the mean trap catch of this species was equal in depot and non-depot traps. In contrast, *T. castaneum* was more frequently trapped in depot traps than non-depot traps during the colder months, but was much more widespread across the landscape during the summer months. *Tribolium castaneum* also showed a clear mid-summer peak in flight activity, whereas *R. dominica* flight activity was highly variable throughout the study. In general, our results reveal patterns that are consistent with those found for *T. castaneum* and *R. dominica* in southern Queensland. The contrasting spatio-temporal patterns of flight activity of *R. dominica* and *T. castaneum* show that species-specific approaches may be needed to manage these pests and that the spatio-temporal dynamics of resistance genes may differ across these species.

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1. Introduction

The rust-red flour beetle, *Tribolium castaneum* (Herbst), and the lesser grain borer, *Rhyzopertha dominica* (F.), are serious pests of stored grain in many countries, and they commonly occur together in infestations (White, 1988; Vela-Coiffier et al., 1997; Athanassiou et al., 2005; Flinn et al., 2010). The threat they pose to stored grain is compounded by their potential to develop resistance to the fumigant phosphine (Jagadeesan et al., 2012; Mau et al., 2012a,b, Opi et al., 2012) and grain protectant insecticides such as deltamethrin (a synthetic pyrethroid) and methoprene (an insect growth

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regulator) (Collins, 1990; Lorini and Galley, 1999; Daghli et al., 2013). Although *T. castaneum* and *R. dominica* are associated with stored grain, knowledge of how these beetles are distributed across the broader landscape is critical for understanding dispersal from infested grain to un-infested grain, and thus the spread of resistance genes. Farm machinery, such as combine harvesters, can be sources of *T. castaneum* and *R. dominica* beetles colonising newly harvested grain (Sinclair and White, 1980), but colonisation by flying beetles also occurs (Vela-Coiffier et al., 1997; Hagstrum, 2001). The questions that follow include when and how far colonising beetles fly.

It is well known that *T. castaneum* and *R. dominica* beetles fly in the immediate storage environment (e.g. Throne and Cline, 1994), but the availability of synthetic aggregation lures and their use in trapping has greatly facilitated the investigation of the flight of

these two species at much greater spatial scales. This is well-demonstrated by pheromone trapping studies that have provided valuable insights into the field ecology of *R. dominica* in North America, where flying *R. dominica* beetles can be caught near grain storage facilities, in open fields, and also in native vegetation (Edde et al., 2005, 2006; Toews et al., 2006; Mahroof et al., 2010). In one study, the re-trapping of marked *R. dominica* beetles showed that they were dispersing at least 1.6 km from the point of release (Mahroof et al., 2010). There was also strong seasonality in their flight activity, with no beetles captured during the coldest months in Oklahoma, Kansas and Nebraska, when temperatures below 0 °C are common (Edde et al., 2006; Toews et al., 2006). Some have proposed that *R. dominica* overwinters in the USA away from stored grain (Toews et al., 2006) and others have argued that *R. dominica* can develop on resources other than stored grain, but evidence for both is limited (Jia et al., 2008; Mahroof and Phillips, 2012). An alternative explanation for the capture of *R. dominica* beetles away from stored grain is they are trapped while searching for resources in new localities. It is also possible that passive dispersal on winds can occur over large distances, and Fields et al. (1993) suggested that *R. dominica* might be carried to Canada from the USA by this means. Despite the considerable efforts made towards characterising *R. dominica* flight activity in North America, there are no similar published pheromone trapping studies on *T. castaneum*.

In Australia, *T. castaneum* and *R. dominica* were the focus of a study that combined pheromone trapping and gene flow analysis to investigate flight dispersal in a rural landscape of over 7000 km² in inland southern Queensland (Ridley et al., 2011, 2016). *Tribolium castaneum* beetles were trapped near farm silos, in fields, and in native vegetation, although they tended to be trapped in greater numbers near silos (Ridley et al., 2011). Similarly, *R. dominica* beetles were trapped near farm silos, in fields and in native vegetation, but traps near silos tended to catch similar numbers of beetles to field traps (Ridley et al., 2016). Thus, flying *T. castaneum* beetles were aggregated around stored grain, whereas *R. dominica* beetles were flying more widely across the rural landscape. Beetles of both species were caught on every trapping occasion even when mean daily maximum temperatures were 20.5 and 21.5 °C in the winter months of June and July 2009 respectively (though few beetles were caught) (Ridley et al., 2011, 2016). Subsequent population genetics analysis of the trapped beetles using microsatellite markers showed that there was extensive gene flow within each species across the entire study area, and flight dispersal clearly contributed to this (Ridley et al., 2011, 2016).

Here we report the results of a pheromone trapping study in a grain growing district adjacent to the Tropic of Capricorn in Central Queensland, in which we tested the three key conclusions from the earlier pheromone trapping study in southern Queensland (Ridley et al., 2011, 2016) applied to this region: (i) *T. castaneum* and *R. dominica* beetles disperse actively across the rural landscape, (ii) *T. castaneum* beetles tend to be more aggregated around stored grain, and (iii) flight activity in both species occurs year-round.

2. Materials and methods

2.1. Study area

The study was undertaken in a humid subtropical grain growing district adjacent to the Tropic of Capricorn in the Central Highlands of Queensland, Australia. Long-term records for the nearest town, Emerald, show that July is the coldest month with a mean minimum of 9.0 °C and a mean maximum of 23.2 °C, and January is the hottest with a mean minimum of 22.2 °C and a mean maximum of 34.4 °C (Australian Bureau of Meteorology [http://www.bom.gov.au/climate/averages/tables/cw_035264.shtml]). The mean annual

rainfall at Emerald is 553.3 mm with mean monthly rainfall ranging from 18.1 mm in July to 90.2 mm in December. Grain farmers typically grow a winter crop of wheat and a summer crop of sorghum. Harvest is usually from August to November for wheat and from late January to September for sorghum (R. Reid, unpublished data).

2.2. Trapping study

Flight activity of *T. castaneum* and *R. dominica* was monitored along a 28.4 km linear transect running roughly north to south from the outskirts of Emerald. At each of nine sites along the transect we placed a Lindgren four-funnel trap (Contech Inc, Delta, BC, Canada) baited with an aggregation pheromone lure for *T. castaneum* and another for *R. dominica* (Trécé Inc, Adair, OK, USA). Traps were numbered from 1 to 9 with Traps 1 and 9 located at the northern and southern transect ends respectively. Traps 2 and 3 were located at a large grain depot at Emerald (adjacent to a cotton gin) and Traps 7 and 8 were located at a large grain depot at Gindie. These traps were designated 'depot traps', and the remainder 'non-depot traps'. The cumulative distance from Trap 1 (0 km) to each of the remaining traps was 2.4, 3.3, 6.5, 11.9, 18.0, 22.7, 23.0 and 28.4 km for Traps 2–9 respectively. Traps 2 and 3 at the Emerald depot were 0.9 km apart, and Traps 7 and 8 at the Gindie depot were 0.3 km apart. At the time of the study, Traps 1–3 were located on the semi-rural outskirts of Emerald, but there has been considerable urban encroachment since then. To our knowledge, there was no farm-stored grain within 1 km of any trap.

Each trap was suspended from wire spanning two metal pickets so the collection container was about 1.5 m from the ground, and fastened so that movement in wind was negligible. Trapped beetles fell into a collection container containing propylene glycol as a preservative. We trapped for 1 year beginning on 15 March 2011 with lures replaced and any trapped beetles collected at 2-wk intervals (fortnightly). There is no published information on longevity of the *T. castaneum* lure, but trapping in the USA suggests that the *R. dominica* lure releases pheromone for at least 2 weeks, with more released during the first than the second week (Toews et al., 2006). Only four (1.7%) of the planned 234 samples could not be collected, because of grass fire (2 samples) or spillage (2 samples). Samples collected from traps were transferred to Brisbane where the numbers of *T. castaneum* and *R. dominica* adults were counted.

Meteorological data recorded for the Emerald Airport were sourced from the Bureau of Meteorology. Laboratory studies using constant temperatures provide lower flight threshold estimates of 25 °C for *T. castaneum* (Cox et al., 2007; Perez-Mendoza et al., 2014) and 19.9 or 20 °C for *R. dominica* (Dowdy, 1994; Cox et al., 2007). Both *T. castaneum* and *R. dominica* tend to fly late in the day, before sunset (e.g. Boon and Ho, 1988; Wright and Morton, 1995), so we used daily maximum temperature as an approximation for temperature at time of peak flight. We then determined the percentage of days during the study in which the maximum temperature fell below 25 and 20 °C.

2.3. Statistical analyses

Trap catch was analysed separately for *T. castaneum* and *R. dominica* using repeated measures ANOVA (GenStat, 2008), to investigate the effects of trapping date and trap location (i.e. depot or non-depot). The data were transformed ($\log_e(N + 1)$) before analysis to conform to the assumptions of ANOVA, and blocked according to trap location (i.e. depot or non-depot). Mean separation was calculated with Fisher's Protected LSD. Non-parametric correlation (Spearman's rank coefficient) was used to test the

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