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Factors Influencing Flight Capacity of the Mountain Pine Beetle (Coleoptera: Curculionidae: Scolytinae)

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Abstract

The mountain pine beetle, *Dendroctonus ponderosae* Hopkins (Coleoptera: Curculionidae: Scolytinae) is the most damaging pest of mature pine (Pinaceae) in western North America. Although mountain pine beetles have an obligate dispersal phase during which adults must locate a new host for brood production, dispersal is a poorly understood aspect of its ecology. This flight mill study was designed to test the effects of beetle size, sex and age on flight capacity. Energy use during flight was assessed through measurements of weight before and after flight and fat content of flown *versus* control beetles. The mean flight distance achieved by mountain pine beetles varied between 2.12 and 5.95 km over the 23 h bioassay but the longest total flight of an individual beetle was > 24 km. Beetle pre-flight weight influenced flight initiation, flight distance and duration. Bigger beetles are more likely to fly and once in flight fly longer and farther than smaller beetles. There was no direct effect of beetle sex on flight capacity. Flight capacity of beetles declined with age post emergence. Although individual flight capacity was variable, flight velocity was relatively constant between 1.55 and 1.93 km/h. Lipids are used to power flight in mountain pine beetles, as lipid content was lower in beetles flown on the flight mills compared to beetles that did not fly. Flight distance was negatively correlated with beetle post-flight lipid content. The baseline flight capacity data revealed in this study have implications for understanding the population dynamics of this eruptive forest pest.

Key Words: dispersal, flight, lipid, bark beetle, *Dendroctonus ponderosae*

The mountain pine beetle, *Dendroctonus ponderosae* Hopkins (Coleoptera: Curculionidae: Scolytinae), is the most damaging pest of mature pine (Pinaceae) forests in western North America (Safranyik and Carroll 2006). Epidemic mountain pine beetle populations have killed an estimated 18.3 million ha of lodgepole pine (*Pinus contorta* Dougl. ex. Loud.) in British Columbia during the most recent widespread outbreak in western Canada (www.for.gov.bc.ca). This outbreak generated an unparalleled source population that produced massive long-distance dispersal events and subsequent range expansion of the beetle into pine forests east of the Rocky Mountains in Alberta (Safranyik et al. 2010), where it now threatens the boreal forest (Cullingham et al. 2011). In the current epidemic, the mountain pine beetle has rapidly adapted to novel habitats at higher latitudes (Cudmore et al. 2010) and altitudes (Mitton and Ferrenberg 2012) that contain novel pine hosts (Cullingham et al. 2011).

Despite extensive research in the native range of the mountain pine beetle, dispersal remains a poorly understood aspect of its ecology and one that is crucial to understand as warming climate conditions permit range expansion (Safranyik et al. 2010). Bark beetles have an obligatory dispersal phase in which adults need to locate new hosts for brood production. Mountain pine beetle movement within and between infested patches and to new uninfested patches can include stand-level dispersal (Safranyik et al. 1992, Robertson et al. 2007) and long distance dispersal above the forest canopy aided by wind (Jackson et al. 2008, de la Giroday et al. 2011, 2012). As only 0.2% of beetles in a population are estimated to disperse above the canopy (Safranyik et al. 1992), the dominant dispersal method is within-stand dispersal by flight at mid-bole height (Safranyik et al. 1989).

Females are the pioneering sex in the host location and colonization process for mountain pine beetles. Upon reaching a suitable host tree (Pureswaran and Borden 2005), females release an aggregation pheromone that attracts both sexes of mountain pine beetle for mass attack of the tree (Pitman et al. 1968). As host colonization proceeds, arriving males produce a different aggregation pheromone that attracts mainly females until the attack density dictates emission of anti-aggregation pheromones by both sexes to prevent further colonization of the host tree (Rudinsky et al. 1974, Ryker and Libbey 1982). In stand-level movement between host trees, mountain pine beetles often disperse past suitable hosts as shown by spatial-temporal analyses of current and past year beetle infestation (green: red attacked trees). This could indicate that variation in beetle dispersal is dependent in part on individual beetle physiology and not solely on the number and distribution of susceptible hosts in a stand (Robertson et al. 2007).

Individual variation in flight capacity by *Dendroctonus* bark beetles has been linked to beetle physiological characteristics in some species. The Chinese white pine beetle, *D. armandi* Tsai and Li, requires carbohydrates for survival and flight initiation but lipid for sustained flight. The lipid content of *D. armandi* is related to the total distance and duration of flight as measured in flight mill bioassays (Chen et al. 2011). The lipid content of Douglas-fir beetles, *D. pseudotsugae* Hopkins, dictates the patterns of adult behavior. *D. pseudotsugae* that contain 10% or less body lipid content do not readily fly whereas, those with greater than 20% body lipid content are not immediately responsive to host cues and must disperse by flight. *Dendroctonus pseudotsugae* with intermediate lipid content are good fliers and are capable of response to volatile cues released by their host trees (Atkins 1966). Evidence that lipid is used to fuel flight in bark beetles comes from studies in which fat is extracted from beetles that have flown on flight mills in the laboratory and compared to similar control beetles that are not given the opportunity to fly (Atkins 1969, Thompson and Bennett 1971, Jactel 1993, Kinn et al. 1994, Williams and Robertson 2008, Chen et al. 2011). The body lipid content of *D. pseudotsugae* that were flown on flight mills is lower than similarly sized control beetles (Atkins 1969). After 5 h of flight, male *D. pseudotsugae* fat content decreased from 14.79% to 5.21% due to selective oxidation of monounsaturated fatty acids (Thompson and Bennett 1971). Both male and female southern pine beetles, *D. frontalis* Zimmerman, metabolize lipids during flight and the amount of fat remaining post flight is negatively correlated with flight duration (Kinn et al. 1994).

Despite the importance of body size and lipid content on *D.* bark beetle flight capacity, there is not always a difference in flight capacity between the sexes. Although female *D. armandi* are larger and have a higher fat content than males, flight capacity as measured by flight distance and duration is similar between the sexes (Chen et al. 2010, 2011), but females are capable of longer sustained flight than males (Chen et al. 2011). Flight distance and duration decrease with age post emergence in both male and female *D. armandi* unless beetles are provided with a phloem nutrition source (Chen et al. 2011). Male and female *D. frontalis* have a similar flight velocity but females fly for longer and cover a greater distance than males. Sustained flight of *D. frontalis* measured in flight mill bioassays lasts for an average of 169.12 and 143.22 minutes during which 3370.7 and 2706.0 m are flown, by female and male beetles, respectively (Kinn et al. 1994).

The objective of this study is to obtain baseline data on the flight potential of mountain pine beetles. The study is designed to assess the effect of beetle size, sex and age on flight potential. Energy use during flight is also assessed by measurement of weight loss during flight and fat content of beetles that have flown compared to similarly treated control beetles that are not provided the opportunity to fly. The findings of this manuscript are intended to lay the foundation for future studies of beetle dispersal under more natural conditions.

Materials and Methods

Beetles. In October 2011, two *D. ponderosae*-infested lodgepole pine trees were felled at each of three different sites near Grande Prairie, Alberta, Canada (54.69° N, 119.02° W). One 50 cm bolt from 1 m above the soil surface was cut from each of the six trees. Bolts were transported to the laboratory at the University of Alberta and the cut ends were sealed with paraffin wax to prevent desiccation before storage at 5°C until use. In January 2012, the six bolts were placed at room temperature (24°C) in separate 121 L bins made of opaque plastic and fitted with emergence jars. Beetle emergence was monitored daily and collected beetles were separated by sex (Lyon 1958). Emergent beetles were placed individually in 2 ml microcentrifuge tube with a small piece of paper and stored at 4°C until use in the flight bioassay. These conditions were used to minimize metabolism of stored resources during the aging process (Elkin and Reid 2005).

Beetle Flight. Differently aged beetles of both sexes were used in flight capacity trials to determine the effect of beetle sex and age on flight. Beetles were removed from cold storage and prepared for flight at different times post emergence as young (1-3 d), middle-aged (5-7 d) or old (9-11 d) beetles. This range of age groups was chosen because most mountain pine beetle dispersal occurs within the first 3 d post emergence in the field (Safranyik et al. 1992) and beetles are considered to be sexually mature by 11 d post emergence (Safranyik and Carroll 2006). Male and female beetles were flown on alternate days. Individual beetles were weighed to the nearest 0.01 mg (Mettler Toledo, XS105, Columbus, OH) and then tethered by attachment to a 0.03 mm diam. aluminum wire with a 0.14 mm diam. loop and a 2 cm straight portion of wire perpendicular to the loop (Fig. 1A). The loop was attached to the beetle pronotum with Pres-Tite Contact Cement (LePage, Mississauga, ON, Canada), so that elytra movement was not impeded. Between 1 and 21 beetles were tethered on each of 40 d. On each of the 40 test days, a random sample of the tethered beetles were not flown but were kept as control beetles. Control beetles were tethered in the same manner as the flown beetles, but instead of being attached to the flight mill, the tether was removed and the beetles were positioned in perforated microcentrifuge tubes (0.2 ml) and held in the flight mill room under the same conditions as the flown beetles for the duration of the bioassay. Tethered beetles were transferred to the chamber housing the flight mills that was maintained at 24°C under a LD 16:8h photoperiod. Each wire harness was attached to the distal end of a flight mill arm at ~100° with the mill arm by a 3 cm length of wire insulation glued to the mill arm (Fig. 1B and 1C). Light (621 lux) was provided by high flicker frequency fluorescent bulbs to avoid interference with beetle behavior (Miall 1978). Flight assays were initiated 4 h after the beginning of the light phase. Each rotation of the mill arm was detected by a small magnetic transmitter/receiver situated on the mill casing that registered a signal to the attached computer. The computer recorded each revolution (94.2 cm) as beetles propelled the arm. The output from the software (LabView™, National Instruments Corporation, Austin, TX) included total distance flown, longest single flight, flight duration and flight speed. At the end of the 23-h flight period, beetles were removed from the flight mills, the tether detached and the beetles were weighed again and stored at -20°C for future lipid extraction. Weight loss during flight was calculated as the difference in weight before and after flight. The difference in weight before and after flight was converted into a proportion by dividing the weight difference by the pre-flight weight. Beetles that were dead at the end of the 23 h flight period were not included in subsequent analyses, which were conducted on 148 control and 174 flown beetles.

Lipid extraction. The energetic condition of flown and control beetles was assessed by lipid extraction (Atkins 1969) using petroleum ether (Fisher Chemical, Fair Lawn, NJ). Beetles were dried in an oven at 60°C for 24 h, and then weighed to the nearest 0.01 mg. Beetles were placed individually into perforated 0.2 ml microfuge tubes and submerged in petroleum ether in a Soxhlet apparatus (45/50 Pyrex; Fisher Scientific, Canada). Lipid was extracted for 8 h. After extraction, beetles were again dried at 24°C for 24 h and then weighed. Lipid mass remaining after flight was calculated as the difference in dry weight before and after extraction. Lipid mass after flight was converted into a relative measure by dividing by pre-extraction beetle dry weight.

Statistical analyses. All data analyses were conducted in R v. 3.0.0 2013-04-03 (R Core Development Team 2013). In all analyses, model simplification was achieved through removal of non-significant factors based on ANOVA hypothesis testing of full and reduced models and comparison of AIC values for model performance, until the most parsimonious model remained.

The propensity for flight of mountain pine beetles measured on the flight mills was analyzed using a logistic regression model. Pre-flight weight, beetle sex, beetle age were specified as independent variables. The original model included all two-way interactions among independent variables. The bolt beetles were reared in was specified as a blocking factor.

Flight capacity was analyzed with general mixed effects models that specified total flight distance, longest single flight, flight duration and flight velocity as dependent variables in each model, respectively. Dependent variables were log transformed prior to analyses to improve normality. Fixed factors in all models were specified as pre-flight weight, beetle sex, and beetle age with tree bolt treated as random factor. Original models included all two-way interactions among fixed factors. Significant effects for factors with more than two levels were subsequently compared using Tukey-Contrasts. As pre-flight weight was determined to be an important factor that influences the flight capacity of beetles, an additional general mixed effects model tested factors that could affect pre-flight weight. Fixed factors were specified as beetle sex, and beetle position in the emergence period and tree bolt was treated as a random factor.

The energetic condition of experimental beetles was assessed in five separate general mixed effects models. The proportional and total body lipid content remaining after the bioassay was compared between flown and control beetles in separate models with fixed factors specified as beetle sex, beetle age, and flight treatment (flown or control) with tree bolt from which beetles emerged treated as a random factor. Original models included all two-way interactions among fixed factors. The effect of flight distance on energy use was assessed only for beetles that were exposed to the flight treatment. A general mixed effects model specified the proportional fat loss during flight as the dependent variable and beetle sex, age, and the total distance flown as fixed factors. The tree bolt from which beetles emerged was treated as a random factor. The original model included all two-way interactions among fixed factors. The proportional and total weight loss during flight was assessed only for beetles that flew (n=174). General mixed effects models specified the proportional and total weight lost during flight as the dependent variables and beetle sex, age and pre-flight weight as fixed factors. The tree bolt from which beetles emerged was treated as a random factor. Two-way interactions of all fixed factors were included in the original models.

Results

Beetle Propensity to Fly. Most of the beetles tethered to flight mills in this study flew. Between 83 and 91% of the tested beetles flew in the different age*sex combinations (Table 1). Beetle propensity to fly was significantly influenced by the pre-flight weight of beetles ($\chi^2=34.95_{1,194}$ $P<0.0001$). Heavier beetles were more likely to fly than light beetles. Despite the fact that female beetles are heavier than males ($F_{1,290}$=98.10 P<0.0001), there was no significant impact of sex on flight propensity (χ^2=0.60$_{1,197}$ P=0.4374). The bolt that beetles were reared in had a marginally significant effect on whether or not they took flight (χ^2=9.97$_{5,189}$ P=0.0760), suggesting that developmental conditions of juvenile beetles may impact dispersal capacity of adults.

Beetle Flight Capacity. Pre-flight weight significantly impacted the total distance flown ($F_{1,164}$=23.46 P<0.0001), the longest single flight ($F_{1,164}$=21.71 P<0.0001), and the duration of flight ($F_{1,162}$=19.60 P<0.0001) of adult beetles. Although there was large variation in the distance and duration of flight, heavier beetles were able to fly farther and for longer than light beetles over the 23 h flight period (Fig. 2). Despite the finding that female beetles are significantly heavier than male beetles, there was no direct effect of beetle sex on total flight distance ($F_{1,164}$=2.12 P=0.1477), longest single flight ($F_{1,164}$=1.68 P=0.1963), and only a marginally significant effect of sex on flight duration ($F_{1,162}$=3.41 P=0.0666) (Table 1).

Beetle age post emergence significantly impacted the total distance flown by beetles ($F_{2,164}$=7.02 P=0.0012). Old beetles that were flown 9-11 d post emergence flew a shorter total distance than either young or middle-aged beetles that were flown 1-3 or 5-7 d post emergence (Fig. 3A). The longest single flight conducted by beetles was also affected by beetle age ($F_{2,164}$=4.70 P=0.0104). Young beetles were able to fly continuously in a significantly longer single flight than old beetles. The longest single flight conducted by middle-aged beetles was intermediate between that of young and old beetles (Fig. 3B). There was a significant overall effect of beetle age on flight duration ($F_{2,162}$=4.22 P=0.0163). Young beetles flew longer than old beetles and flight time by middle-aged beetles was intermediate (Fig. 3C).

The interaction between beetle pre-flight weight and beetle age significantly influenced beetle flight velocity ($F_{1,141}$=3.46 P=0.0340). Although flight velocity of beetles is quite constant, a few light beetles flew very fast (Fig. 4). Fast flight by light beetles in the older age categories appears to be driving this relationship (Fig. 4).

Beetle Energetic Condition. As would be expected, beetles that flew had a smaller proportion of body lipid content remaining after the bioassay than control beetles that were not given the opportunity to fly ($F_{1,305}$=91.24 P<0.0001) (Fig. 5). Females had a greater proportion of body lipid content than males ($F_{1,305}$=28.76 P<0.0001). There was no main effect of beetle age on the proportional ($F_{2,305}$=0.18 P=0.8356) or absolute ($F_{2,304}$=0.38 P=0.6818) body lipid content of beetles but an interaction effect of sex*age influenced proportional ($F_{2,305}$=2.95 P=0.0541) and absolute ($F_{2,304}$=3.71 P=0.0056) beetle body lipid content (Fig. 6). Young and medium-aged female beetles had a greater proportional and absolute body lipid content than similarly aged males whereas, body lipid content was similar between the sexes in old beetles (Fig. 6). Absolute fat content of beetles measured after the bioassay was affected by a significant interaction between sex and flight treatment ($F_{1,304}$=7.7894 P=0.0056). The difference in fat content between the sexes was greater for control than flown beetles (Fig. 7) suggesting that males and females metabolize fat for fuel differently than males during flight.

Total distance flown also influenced the proportional ($F_{1,141}$= 11.75 P=0.0008) and absolute ($F_{1,141}$= 8.20 P=0.0048) body lipid content measured after flight. The proportion of body lipid content of beetles measured after flight decreased with a total distance flown (Fig. 8), suggesting that fat is the main fuel for mountain pine beetle flight. Females had a higher proportional ($F_{1,141}$=8.73 P=0.0037) and absolute ($F_{1,141}$=18.74 P<0.0001) body lipid content remaining after flight than males.

Pre-flight weight significantly affected the proportional ($F_{1,164}$=7.74 P=0.0060) and absolute ($F_{1,162}$=85.03 P<0.0001) weight lost during flight. A significant interaction between beetle age and pre-flight weight influenced the absolute weight lost ($F_{2,162}$=3.47 P=0.0335) during flight (Fig. 9). As would be expected, absolute weight lost during flight increased with beetle weight but the rate of increase differed with beetle age (Fig. 9). Females lost more absolute weight during flight than males ($F_{1,162}$=91.82 P<0.0001) but beetle sex had no effect on the proportional weight lost as a result of flight ($F_{1,164}$=2.45 P=0.1193).

Discussion

This study elucidates some of the important factors that influence mountain pine beetle dispersal by flight. Controlled flight experiments using flight mills reveal individual variation in flight capacity among beetles that provides baseline data on beetle dispersal under laboratory conditions. The study captured a wide range of variation in flight capacity, as between 83-91% of the mountain pine beetles that were tethered to the flight mills initiated flight (Table 1). This is in contrast to other species of bark beetles in which a large portion of the test population does not initiate flight when positioned on a mill (Atkins 1959, Atkins 1966, Jactel 1993, Kinn et al. 1994, Robertson and Roitberg 1998). Approximately, 33% of *Ips sexdentatus* Boern do not fly in flight mill bioassays in the laboratory (Jactel 1993).

The mean flight distance achieved by mountain pine beetles varied between 2.12 and 5.95 km (Table 1) over the 23 h bioassay but the longest total flight of an individual beetle was over 24 km. These distances are comparable to those achieved by the congener, *D. frontalis*, that flies on average between 2.7 and 3.4 km during flight mill bioassays (Kinn et al. 1994). The large inter-individual variation in flight capacity of mountain pine beetles revealed in this study also occurs in *D. armandi* that fly between 0.0003 and 48.9 km on laboratory flight mills (Chen et al. 2010). Measurements of flight of *D. pseudotsugae* on laboratory flight mills suggest that beetles are capable of longer flights between 24-32 km in a single day (Atkins 1961) but more recent work shows average flight distances of 4.6 km for this species (Williams and Robertson 2008). Flight distances of mountain pine beetles measured under field conditions in response to semiochemical-baited traps are mainly less than 30 m (Safranyik et al. 1992). This suggests that flight capacity of bark beetles measured on flight mills may be an over estimate because the beetle is supported by the flight apparatus (Williams and Robertson 2008) or that short range dispersal is curtailed under field conditions in the presence of attractive semiochemicals or as a result of environmental conditions that are suboptimal for beetle flight.

Beetle pre-flight weight influences both flight initiation and flight distance and duration by mountain pine beetles. Bigger beetles are more likely to fly and once in flight fly longer and farther than smaller beetles. However, energy use also increases with body size as weight lost during flight increased with the size of flying mountain pine beetles. The effect of body size on flight capacity in bark beetles varies depending on the species and life history of the beetle. Body size is positively related to dispersal capacity in bark beetles that show sexual size dimorphism as adults (Atkins 1966) in which females are the pioneering sex (Kinn et al. 1994), like many *Dendroctonus* species. Pre-flight weight is correlated with flight duration and distance in *D. frontalis* (Kinn et al. 1994) but only duration in *D. armandi* (Chen et al. 2011). Body size is a less important determinant of dispersal capacity in bark beetles in which males are the pioneering sex and adults do not display sexual size dimorphism, like many *Ips* species. Despite the small body size of *I. sexdentatus*, adult beetles are capable of long distance flight that measures between 5 and 50 km on laboratory flight mills (Jactel and Gaillard 1991). There is no correlation in flight capacity and body size in *I. sexdentatus* (Jactel 1993). Similarly, flight mill studies of *I. typographus* L. reveal that flight distance and duration are independent of body size (Forsse and Solbreck 1985). This is supported by mark-recapture studies in which similarly sized *I. typographus* are captured at various distances from the release location. Body size may influence the direction of dispersal by *I. typographus* as larger male beetles are more likely to disperse upwind from the release location than smaller males (Botterweg 1982). However, body size does influence flight capacity of the pine engraver beetle, *I. pini* (Robertson and Roitberg 1998). Large male *I. pini* positioned on a flight mill in the laboratory fly significantly faster for a greater distance than small males.

This study clearly shows that body size is an important determinant of dispersal capacity of mountain pine beetles. Although mountain pine beetle adults are sexually size dimorphic, sex did not directly influence flight capacity. This may be due to the large variation in flight capacity as measured by flight distance and duration in this study. In other sexually size dimorphic bark beetles like *D. frontalis*, the larger females fly for longer time periods over greater distances than males (Kinn et al. 1994). There is no difference in overall flight capacity between sexes in *D. armandi* during the first 24 h flight period but larger females are capable of longer sustained flight than males during repeated testing (Chen et al. 2011). A longer sustained flight might be adaptive for pioneering females in search of suitable host trees at distances from the natal host tree. Based on the current study, there is no evidence that flight capacity differs by sex in mountain pine beetles. It is possible that a difference in sustained flight capacity would have been observed between the sexes if beetles were allowed to fly for more than 23 h. As female pioneer mountain pine beetles randomly land on potential host trees and assess host suitability at close range (Pureswaran and Borden 2005), dispersal in nature may consist of several flights rather than long periods of sustained flight. The lack of difference in flight capacity between male and female mountain pine beetles may be due to a difference in lipid metabolism between the sexes. Females had more lipid stores than males in the young and middle-aged groups but lipid levels were similar between the sexes in old beetles, suggesting that females use fat for survival to a greater degree than males. In other bark and ambrosia beetles, fat content is generally greatest in the sex that is the pioneer for host colonization. In species in which the female is the pioneer, such as *D. frontalis* (Kinn et al. 1994), *D. armandi* (Chen et al. 2011) and *T. lineatum* (Nijholt 1967) females have more fat than males. Males have higher lipid stores in species in which the males are the pioneers (Slansky and Haack 1986) but this does not always relate to differences in flight capacity between the sexes. Male and female *I. sexdentatus* have similar dispersal capacities and it is hypothesized that only a threshold level of fuel is necessary to initiate flight in this species (Jactel 1993).

Dispersal capacity of mountain pine beetles declined with age post emergence. Flight duration and distance was lowest for beetles that were flown at the oldest age category, 9-11 d post emergence (Table 1). Similarly, *Ips calligraphus* (Germar) flew the farthest 2 d after emergence and flight capacity decreased to zero by 9 d post emergence that were flown repeatedly between 1 and 4 d post emergence (Chen et al. 2011). The difference in flight capacity by the variously aged beetles in the current study may be due in part to reduced lipid stores in older female beetles as a result of metabolism during storage. However, lipid levels were similar across ages in male beetles suggesting that the effect of starvation on lipid metabolism varies between the sexes and could differentially affect dispersal. This finding suggests that starvation may not be a good surrogate of energy use during flight, as has been assumed in previous studies (Elkin and Reid 2005). Although access to a phloem resource may offset the effect of beetle age on flight capacity as in *D. armandi* (Chen et al. 2011), mountain pine beetles are not known to feed post emergence from the natal tree prior to dispersal (Elkin and Reid 2005). Therefore, the effect of beetle age on flight capacity illustrated in this study probably accurately reflects age effects on dispersal under field conditions. Over 80% of released newly emerged mountain pine beetles were captured at trap trees within 3 d of release in a mark-recapture study (Safranyik et al. 1992).

Despite wide variation in the distance and duration of flight by mountain pine beetles, flight velocity was relatively constant ranging between 1.55 and 1.93 km/h (25.83 and 32.17 m/min; 0.43 and 0.54 m/s) (Table 1). Flight velocity is also relatively constant in other species of bark beetle. Although female *D. frontalis* are capable of flying longer distances for longer time periods than males, the velocity of flight does not differ between the sexes (Kinn et al. 1994). The flight velocities measured in the current study are similar to those of other *Dendroctonus* species measured on flight mills. The mean flight velocity of *D. pseudotsugae* (Williams and Robertson 2008) and *D. frontalis* (Kinn et al. 1994) measured in similar assays is 0.36 m/s and 19.22 m/min, respectively. The flight velocity of mountain pine beetles measured in the current study is lower than the estimate of ~2 m/s for this species from field studies (Safranyik et al. 1989). It is unclear how the velocity of beetle flight in the field was estimated (Safranyik et al. 1989) but this discrepancy could mean that flight speed is impaired by attachment to the flight apparatus under lab conditions.

Flight of the mountain pine beetle is fuelled, at least in part, by lipid metabolism. Beetles subjected to flight treatment in flight-mill bioassays contained less fat than similarly treated beetles that were not provided the opportunity to fly. Dissections of adult female mountain pine beetles on emergence from the natal host tree support these conclusions. Emergent female beetles about to embark on dispesal flights have large metathoracic flight muscles and a conspicuous fat body in the metathorax and abdomen. In contrast, females under the bark in the egg laying phase of their life cycle have reduced flight musculature and fat content, suggesting that fat is consumed during the flight and host colonization period (Reid 1958). In the current study, control beetles contained a median 15% of their body mass as lipid; beetles that flew had a median of only 7% of their mass attributed to lipid at the time of measurement. Post emergent *D. frontalis* have an average of 18.2% fat content compared to 10.9% after flight (Kinn et al. 1994). After 5 h of flight, male *D. pseudotsugae* fat content decreased from 14.79 to 5.21% of the body weight (Thompson and Bennett 1971). The importance of lipid as fuel for flight in mountain pine beetles is highlighted by the finding that distance flown on the flight mill was negatively correlated with proportional body lipid content. Lipid stores also decline with increased dispersal by flight in *D. armandi* (Chen et al. 2011), *D. frontalis* (Kinn et al. 1994) and in *D. pseudotsugae* (Thompson and Bennett 1971), *I. paraconfusus* (Hagen and Atkins 1975) and *I. calligraphus* (Slansky and Haack 1986). It appears that the bark and ambrosia beetles studied to date predominately utilize fat to fuel flight musculature for dispersal by flight. The majority of lipid in *D. frontalis* is comprised of triglycerides (Kinn et al. 1994) that is hypothesized to be metabolized to short chain fatty acids during the dispersal and host colonization process (Hodges and Barras 1974). Oxidation of fatty acids by flight muscles powers sustained flight in *D. pseudotsugae* but carbohydrate fuel is important during the initial dispersal process (Thompson and Bennett 1971). A combination of lipids and carbohydrates is used to fuel flight in *D. armandi* in which carbohydrates are important for flight initiation (Chen et al. 2011). Further investigation into the specific fuels used for dispersal by mountain pine beetles is warranted.

The relationships between dispersal capacity of mountain pine beetle with beetle body size and lipid stores uncovered in the current study have implications for understanding the population dynamics of this eruptive forest pest. Bark beetle body size and lipid content vary with development temperature (Nijholt 1967), season (Hedden and Billings 1977), life stage (Hodges and Barras 1974), population density (Williams and Robertson 2008), food source (Chen et al. 2011) and tree host size (Graf et al. 2012). Mountain pine beetle production per tree increases with tree diameter (Safranyik and Carroll 2006) suggesting that the offspring from large hosts disproportionately makes up the population of dispersing beetles. The results of the current study suggest that potential flight distance will be greatest for the largest beetles with the most lipid stores. Levels of stored lipids dictate behavioural patterns in other adult bark beetles. *Dendroctonus pseudotsugae* adults with high levels of stored lipid need to disperse by flight before they become responsive to host cues for colonization (Atkins 1966). This may also be the case in the mountain pine beetle as beetles often disperse past suitable hosts during flight (Robertson et al. 2007). Because the energetic condition of mountain pine beetles is important for other life history traits including host location (Elkin and Reid 2010), colonization (Latty and Reid 2010) and reproductive success (Elkin and Reid 2005), future work will consider the potential for trade-offs between flight capacity and reproduction and host finding behavior of mountain pine beetles.

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Table 1. The effect of sex and age on flight performance of *D. ponderosae*. Values are mean ± SE and sample size is stated in brackets.

	Proportion that flew	Total distance flown (km)	Longest single flight (km)	Total duration of flight (hr)	Total distance flown velocity (km/hr^{-1})	Pre-flight weight (mg)
Female						
Young	0.88	5.53 ± 0.94 (35)	2.86 ± 0.71 (35)	3.22 ± 0.49 (35)	1.75 ± 0.07 (35)	13.81 ± 0.54 (35)
Middle	0.85	5.11 ± 0.97 (33)	2.35 ± 0.52 (33)	2.95 ± 0.47 (33)	1.74 ± 0.08 (33)	14.85 ± 0.48 (33)
Old	0.83	3.14 ± 0.74 (15)	1.23 ± 0.46 (15)	3.18 ± 1.07 (15)	1.55 ± 0.14 (15)	13.44 ± 0.56 (15)
Male						
Young	0.91	5.95 ± 1.05 (29)	3.55 ± 0.87 (29)	3.27 ± 0.54 (29)	1.93 ± 0.12 (29)	10.74 ± 0.47 (29)
Middle	0.88	4.18 ± 0.61 (37)	1.68 ± 0.41 (37)	2.59 ± 0.34 (37)	1.91 ± 0.18 (37)	10.66 ± 0.36 (37)
Old	0.89	2.12 ± 0.61 (25)	1.25 ± 0.54 (25)	1.38 ± 0.38 (25)	1.85 ± 0.29 (25)	10.59 ± 0.37 (25)

Figure Captions

Figure 1. (A) Mountain pine beetle with flight tether affixed to pronotum. Flight mill with tethered beetle attached in (B) side and (C) aerial view.

Figure 2. The relationship between mountain pine beetle pre-flight weight and subsequent flight capacity. (A) Beetle pre-flight weight and total distance flown. (B) Beetle pre-flight weight and total flight duration.

Figure 3. The effect of beetle age post emergence on flight capacity of the mountain pine beetle. (A) Median ± Inter Quartile Range (IQR) total distance flown of variously aged beetles. (B) Median ± IQR longest single flight of variously aged beetles. (C) Median ± IQR total flight duration of variously aged beetles. Whiskers represent data that fall within 1.5 x IQR, while outliers are presented by dots that fall between 1.5-3 x IQR. Within each panel bars labeled with the same letter are not significantly different (Tukey Contrasts, $P>0.05$).

Figure 4. The effect of mountain pine beetle pre-flight weight and age post emergence on flight velocity.

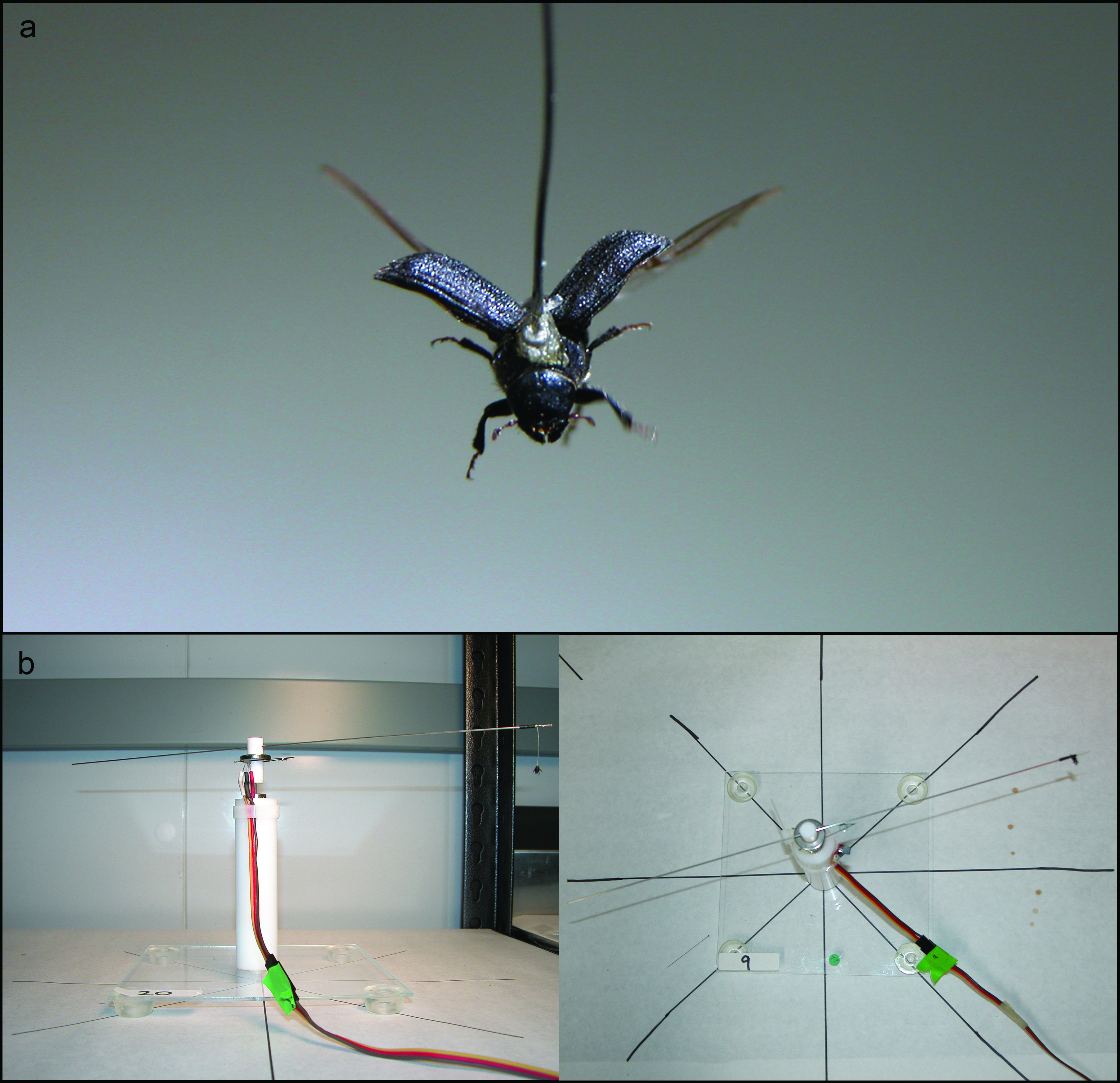
Figure 5. Median ± IQR proportional body lipid content of control vs. flown mountain pine beetles. Whiskers represent data that fall within 1.5 x IQR, while outliers are presented by dots that fall between 1.5-3 x IQR.

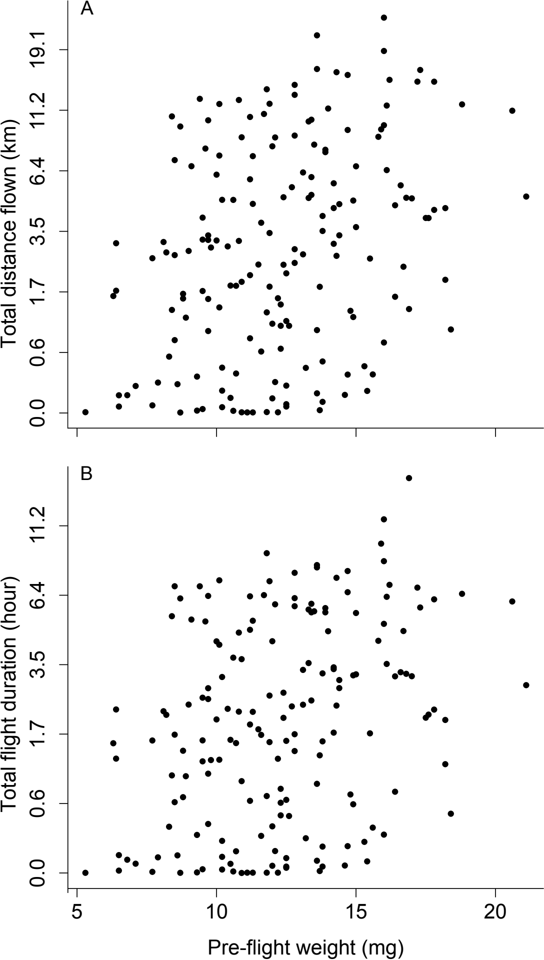
Figure 6. The effect of mountain pine beetle age and sex on beetle body lipid content. (A) Median ± IQR proportional body lipid content. (B) Median ± IQR absolute body lipid content. Whiskers represent data that fall within 1.5 x IQR, while outliers are presented by dots that fall between 1.5-3 x IQR.

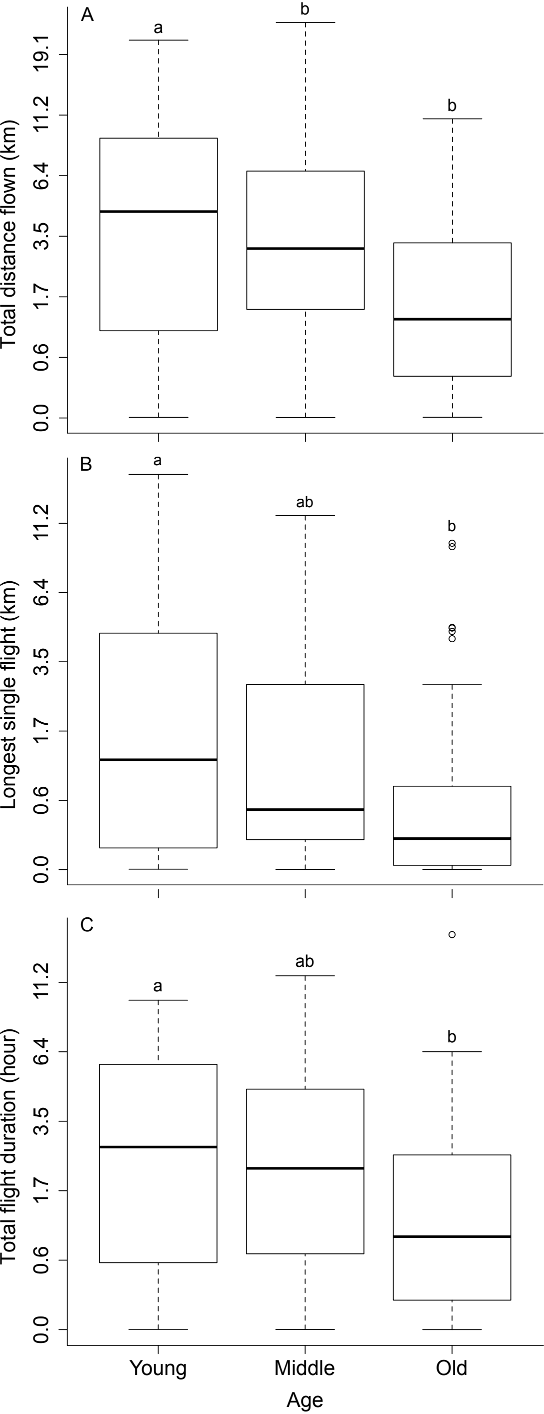
Figure 7. Median ± IQR absolute body lipid content of control vs. flown male and female mountain pine beetles. Whiskers represent data that fall within 1.5 x IQR, while outliers are presented by dots that fall between 1.5-3 x IQR.

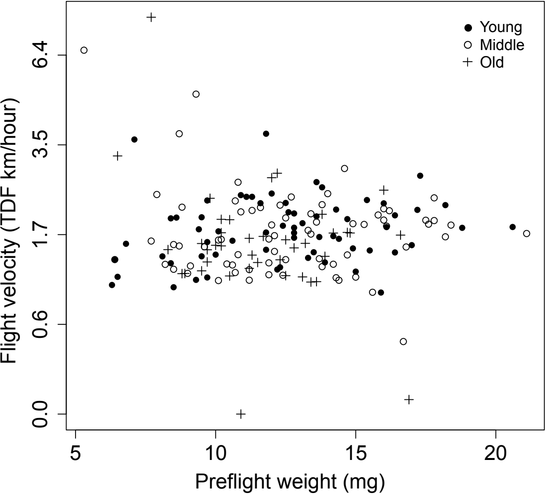
Figure 8. The relationship between the total distance flown by mountain pine beetles and the proportional body lipid content of beetles after the flight bioassay.

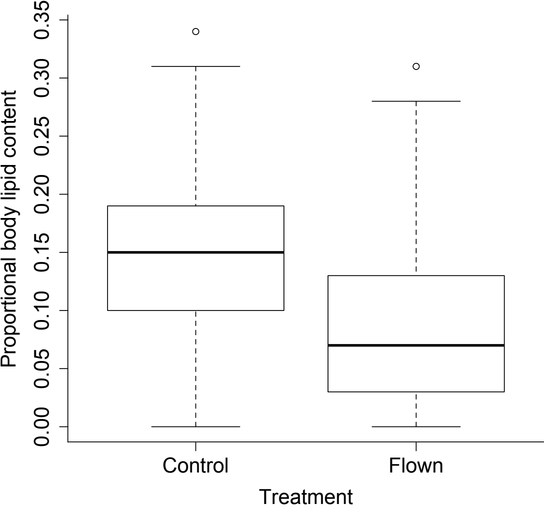
Figure 9. The effect of pre-flight weight of variously aged mountain pine beetles on weight lost during the flight bioassay.

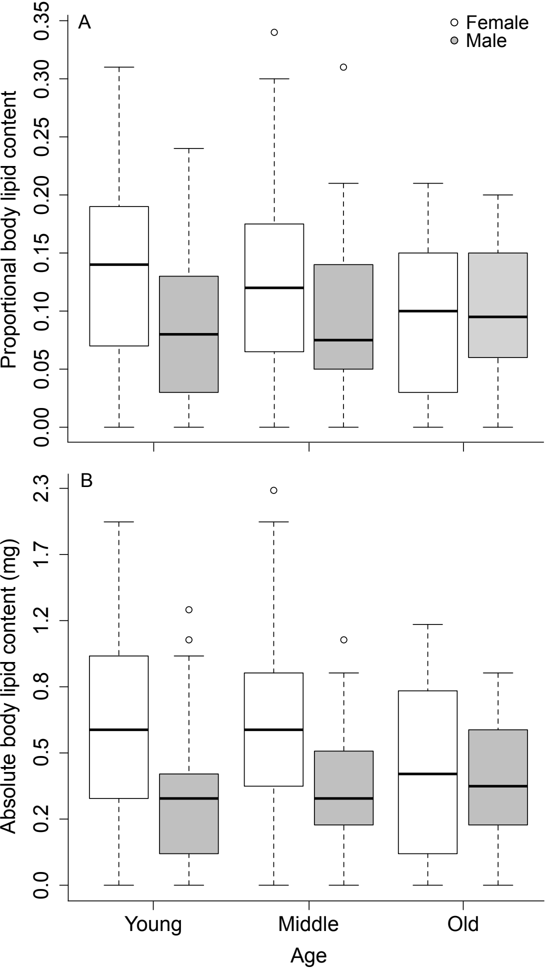

a
b
20
9

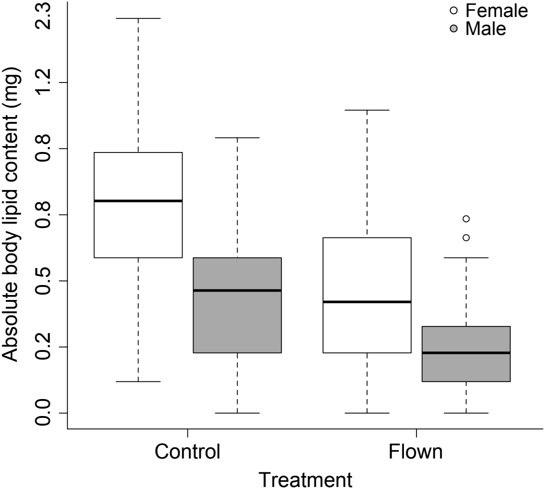

A
Total distance flown (km)
0.0
0.6
1.7
3.5
6.4
11.2
19.1
B
Total flight duration (hour)
0.0
0.6
1.7
3.5
6.4
11.2
5
10
15
20
Pre-flight weight (mg)

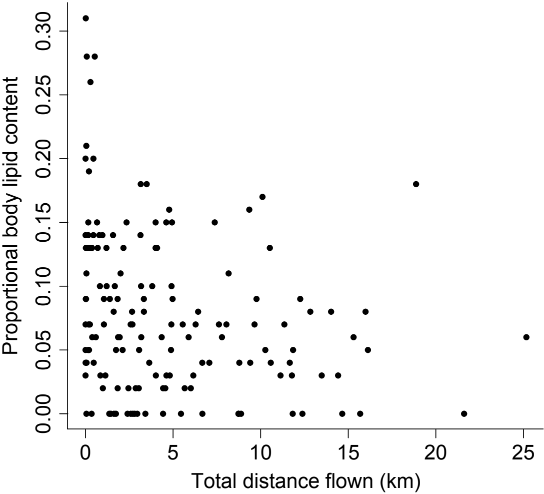

A
Total distance flown (km)
0.0
0.6
1.7
3.5
6.4
11.2
19.1
a
b
b
B
Longest single flight (km)
0.0
0.6
1.7
3.5
6.4
11.2
a
ab
b
C
Total flight duration (hour)
0.0
0.6
1.7
3.5
6.4
11.2
a
ab
b
Young
Middle
Old
Age

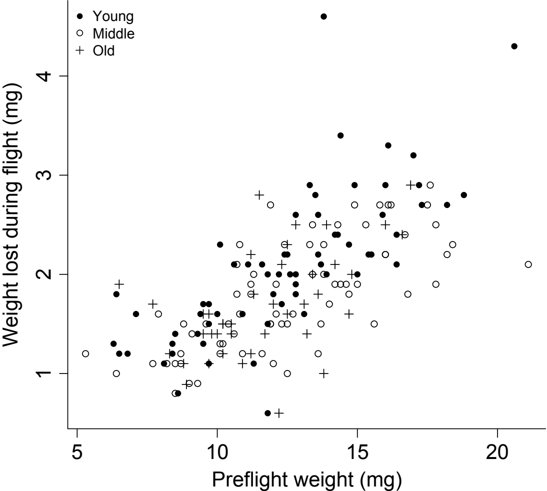

● Young
○ Middle
+ Old
Flight velocity (TDF km/hour)
0.0
0.6
1.7
3.5
6.4
Preflight weight (mg)
5
10
15
20


Proportional body lipid content
0.00
0.05
0.10
0.15
0.20
0.25
0.30
0.35
Control
Flown
Treatment


A
○ Female
● Male
Proportional body lipid content
0.00
0.05
0.10
0.15
0.20
0.25
0.30
0.35
B
Absolute body lipid content (mg)
0.0
0.2
0.5
0.8
1.2
1.7
2.3
Young
Middle
Old
Age


○ Female
● Male
Absolute body lipid content (mg)
2.3
1.2
0.8
0.8
0.5
0.2
0.0
Control
Flown
Treatment


Proportional body lipid content
0.00
0.05
0.10
0.15
0.20
0.25
0.30
0
5
10
15
20
25
Total distance flown (km)


● Young
○ Middle
+ Old
Weight lost during flight (mg)
1
2
3
4
5
10
15
20
Preflight weight (mg)