Age-related activity patterns are moderated by diet in Queensland fruit flies *Bactrocera tryoni*

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Abstract. Life-history parameters and the fitness of tephritid flies are closely linked to diet. Studies of locomotor behaviour can provide insights to these links, although little is known about how locomotor behaviour is influenced by diet. In the present study, video recordings of Queensland fruit flies *Bactrocera tryoni* Froggatt (Diptera: Tephritidae) ('Q-flies') that are maintained individually in cages are used to determine how diet affects the activity patterns (flight, walking, grooming, inactivity) of males and females at ages ranging from 4 to 30 days. The frequency and total duration of activities over 10-min trials are affected by diet, age and sex. Supplementation of diet with hydrolysed yeast results in a higher frequency and duration of flight in flies of all ages and both sexes. The effect of diet on other activities varies with age. Q-flies fed sugar only increase walking frequency steadily from 4 to 30 days post-eclosion, whereas flies fed sugar + yeast have higher walking frequencies at 4 and 10 days than flies fed sugar only, although they then exhibit a sharp decline at 30 days post-eclosion. The frequency and duration of inactivity remain consistent in flies fed sugar + yeast, whereas flies fed sugar only exhibit a marked increase in inactivity from 4 to 30 days post-eclosion. Compared with older flies, 4 day-old Q-flies fed sugar only spend considerably more time grooming. The potential of activity monitoring as a quality control test for flies that are mass-reared for use in sterile insect technique programmes is discussed.

Key words. Behaviour, diet, inactivity, locomotor activity, sugar, Tephritidae, yeast hydrolysate.

Introduction

Nutrition exerts powerful effects on the fitness of many insects (Raubenheimer *et al*., 2009), including tephritid flies. In many tephritid flies, a diet combining sugar with yeast hydrolysate, a rich source of carbohydrates, amino acids, sterols, minerals and vitamins, is found to enhance reproductive development, sexual performance, egg production and lifespan compared with a diet of sugar alone, and is commonly recommended for mass-rearing and pre-release provisioning (Yuval *et al*., 2002, 2007; Liedo *et al*., 2010). Because tephritid flies are important pests, and some species are reared in massive

numbers for release in sterile insect technique programmes, the focus of most nutrition research is on developing dietary regimes that promote rapid development and high reproductive activity. However, to understand the mechanisms by which diet affects these life-history parameters and fitness, we need to understand associated physiology and behaviour. Studies of unrestrained locomotor activity can provide useful insights because behaviour provides a window to physiological state.

Locomotor activity is a central facet of almost all animal behaviour (Martin, 2003) with established links to foraging, reproductive success and predator avoidance (Cobb *et al*., 1987; Oufiero & Garland, 2007; Irschick *et al*., 2008). Being energetically demanding (Lehmann & Schützner, 2010), locomotor activity is expected to interact with diet and associated physiological state, and this expectation is borne out in the limited data that are available for tephritid flies. For

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example, Zou *et al*. (2011) report that daily activity of the Mexican fruit fly *Anastrepha ludens* fed a sugar-only diet is higher than activity of those fed both sugar and yeast extract. However, regardless of diet, the daily total distance travelled by *A. ludens* declines to similar low levels during the second half of the lifespan (Zou *et al*., 2011).

The Queensland fruit fly *Bactrocera tryoni* Froggatt (Diptera: Tephritidae) has been the focus of substantial research into how the inclusion of yeast hydrolysate (and in particular amino acids) in the diet can promote rapid development, high levels of sexual activity and longevity (Taylor *et al*., 2011). The addition of even a small amount of yeast hydrolysate to a diet of sugar increases longevity (Pérez-Staples *et al*., 2007, 2008, 2009; Fanson *et al*., 2009; Fanson & Taylor, 2012a, 2012b).

Despite the substantial attention afforded to the effects of diet on life-history parameters and mating performance, there are no direct studies available regarding how diet affects locomotor activity in *B. tryoni*. A recent study highlights the effects of mass-rearing and irradiation on general activity patterns (flight, walking, grooming, inactivity) in *B. tryoni* (Weldon *et al*., 2010). Mass-reared flies exhibit a marked reduction in overall activity compared with wild-type flies, and reproductive sterilization with gamma radiation (70–75 Gy) is found to induce a further reduction. Unlike many other insects (e.g. *Drosophila melanogaster*: Gatti *et al*., 2000; Hadj Belgacem & Martin, 2002), there is no evidence for sex differences in the general activity of *B. tryoni* (Weldon *et al*., 2010). Drawing on the techniques deployed by Weldon *et al*. (2010), the present study examines the effects of diet on locomotor activity patterns in male and female *B. tryoni* aged 4, 10 and 30 days post-adult eclosion. It is expected that activity patterns will vary with the underlying physiological state as a function of diet, sex and age.

Materials and methods

Source and treatment of flies

Bactrocera tryoni were obtained as pupae from New South Wales Department of Primary Industries mass-rearing facility [Elizabeth Macarthur Agricultural Institute (EMAI), Camden, NSW, Australia]. Further details of the mass-reared colony at EMAI and delivery methods are provided in Pérez-Staples *et al*. (2009) and Weldon *et al*. (2010).

At Macquarie University, *B. tryoni* were maintained in a controlled environment room under a LD 14 : 10 h photocycle at 24–26 °C and 60–70% relative humidity. The illumination regimen consisted of 12 h of full-intensity lighting and simulated dawn and dusk periods in which lights were stepped on and off in four stages over the course of 1 h. Three replicate cohorts of flies from different production periods were used to complete the study.

Adult flies were allowed to emerge from pupae into 5-L ventilated plastic cages (24 × 15 × 15 cm) in the laboratory. The flies were sorted by sex on the day of eclosion, housed in separate 5-L cages (70 flies per cage) and assigned to one of two diet treatments: dry granular cane sugar only, or sugar + yeast (yeast hydrolysate enzymatic, MP Biomedicals, Aurora, Ohio), which was provided in separate dishes to allow self-selection of intake. Flies were provided *ad libitum* access to water and food.

Locomotor activity of female and male *B. tryoni* with a diet of sugar only or sugar supplemented with yeast hydrolysate ('sugar + yeast') was tested at three ages that correspond with recognized stages of sexual development for *B. tryoni* of the tested strain when provided a sugar + yeast diet: 4, 10 and 30 days after adult eclosion. At 4 days after adult eclosion, both female and male *B. tryoni* are still reproductively immature (Meats *et al*., 2004; Pérez-Staples *et al*., 2007). By 10 days after adult eclosion, both females and males are reproductively mature (Pérez-Staples *et al*., 2007). An age of 30 days after adult eclosion is close to the mean longevity of female and male *B. tryoni* when held in single-sex cages (Fanson & Taylor, 2012a). When provided sugar-only diets, *B. tryoni* exhibit low levels of reproductive maturity even after 30 days, and this is especially evident for females (Pérez-Staples *et al*., 2007, 2011; Prabhu *et al*., 2008).

Activity assays

Individual flies were removed from group holding cages and placed in 370-mL clear polypropylene containers (height 47 mm, diameter 119 mm; Chanrol C10, Chanrol Pty. Ltd, Australia), which were fitted with a base of filter paper to provide contrast, and covered with the lid of a clear plastic petri dish (diameter 140 mm). After a 2-min acclimation period, video recordings were made in MPEG-4 format of the activity of individual flies for 10 min periods using a digital video camera (Canon XL1; Canon, Japan) connected through a USB TV tuner (Evolution TV, Model ETV02; Miglia Technology Ltd, U.K.) via an Apple MacBook laptop computer (Apple Inc., Cupertino, California) to an external hard-drive (Maxtor OneTouch III; Maxtor Corporation, Milpitras, California) for storage. Video recordings were made between 10.00 and 16.00 h, with the camera positioned directly above the polypropylene containers, under fluorescent overhead lighting supplemented with two 60-W full spectrum bulbs positioned 500 mm above the plastic containers.

For each replicate, the activity of eight males and eight females from each diet group (sugar only, sugar + yeast) was recorded. The process was repeated at 4, 10 and 30 days post-eclosion, giving 96 recordings per replicate and a grand total of 288 observations over three replicates. Recordings were scored, in real time at a later date, for the frequency and duration of flights, walking, grooming and periods of inactivity using JWATCHER, version 1.0 (Blumstein & Daniel, 2007).

Statistical analysis

Mutually exclusive activities (flight, walking, grooming, inactivity) were compared using multivariate analysis of variance (MANOVA), with diet, sex, age and time of day of recording (morning 10.00–12.00 h; early afternoon 12.00–14.00 h;
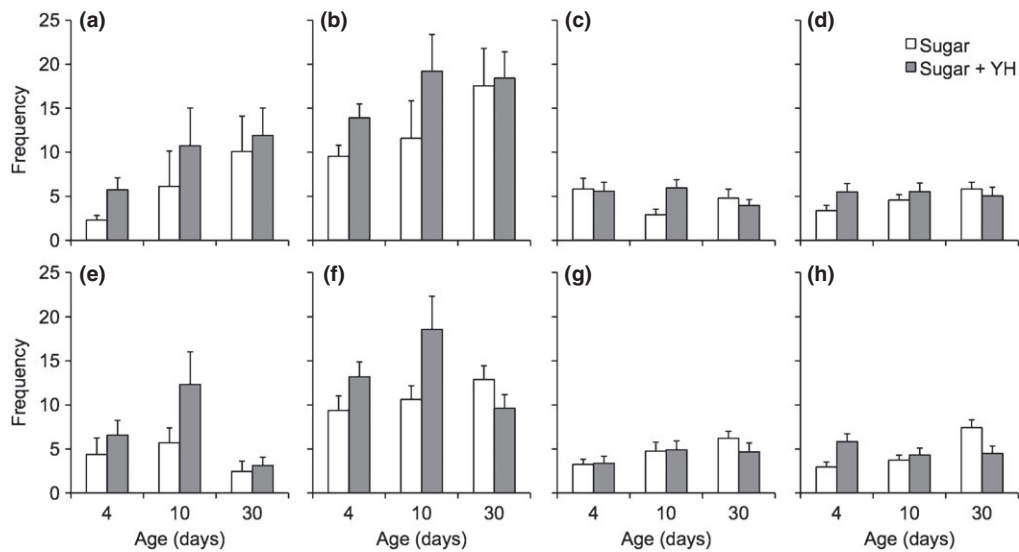
Fig. 1. Frequency of flying, walking, grooming and inactivity in relation to sex, age and diet in fertile mass-reared *Bactrocera tryoni* over a period of 10 min. White bars represent sugar only diets; grey bars represent the sugar + yeast hydrolysate (YH) diet. Frequency of (a) flight, (b) walking, (c) grooming and (d) inactivity in female flies. Frequency of (e) flight, (f) walking, (g) grooming and (h) inactivity in male flies. Raw data are presented as the mean ± SE.

late afternoon 14.00–16.00 h) as fixed effects. Interactions between diet and sex, diet and age, sex and age, diet and time of day, sex and time of day, and age and time of day were also examined. Before the analysis, frequencies and durations of activities were subjected to Box-Cox transformation (appropriate for zero inflated data; Hyndman & Grunwald, 2000) to meet the assumptions of parametric analysis. Arcsine transformations have previously been advocated for this type of data (Fowler *et al*., 1998); however, the practice is no longer recommended (Warton & Hui, 2011). The effect of replicate was discounted in preliminary analysis and this was omitted from the final model (Weldon *et al*., 2010), along with the nonsignificant three-way interactions ($P > 0.1$; Sokal & Rohlf, 1995). We further explored significant effects from MANOVAS in univariate analyses of variance and post-hoc tests of main effects (Tukey's honestly significant difference). All analyses were performed using STATISTICA, version 10 (StatSoft Inc., Tulsa, Oklahoma).
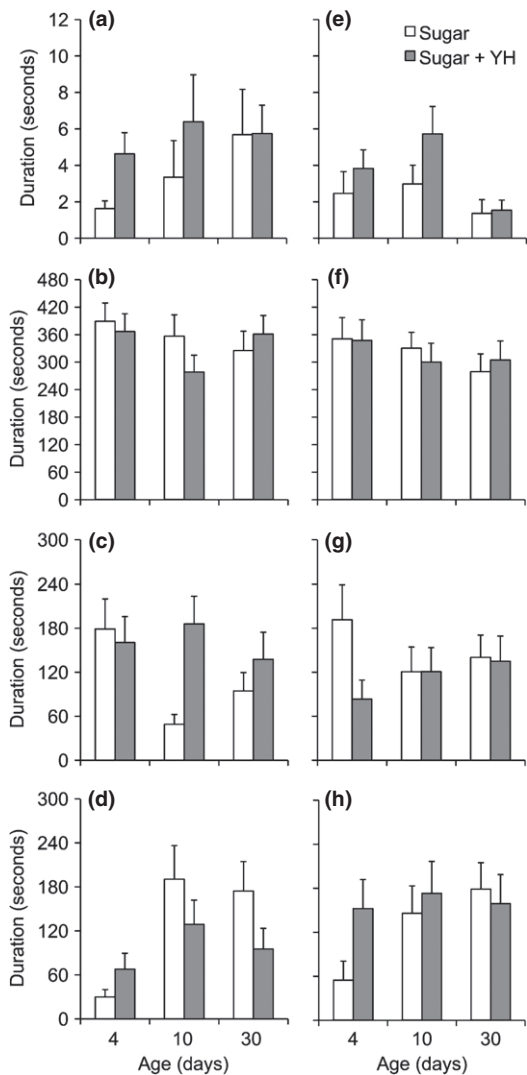
Table 1. Summary of multivariate analysis of variance on the effects of diet (sugar only, sugar + yeast hydrolysate), sex, age (4, 10, 30 days after eclosion), and time of day (ToD; morning 10.00–12.00 h; early afternoon 12.00–14.00 h; late afternoon 14.00–16.00 h) on the frequency of locomotor activity in mass-reared *Bactrocera tryoni* over a period of 10 min, as conducted on Box-Cox transformed data.

Source	d.f.$_{num}$	d.f.$_{den}$	Wilks' λ	F	P
Intercept	4	265	0.1077	548.85	< 0.0001
Diet	4	265	0.9607	2.72	0.0306
Sex	4	265	0.9978	0.15	0.9657
Age	8	530	0.9706	1.00	0.4372
Time of day	8	530	0.9849	0.51	0.8528
Diet × Sex	4	265	0.9869	0.87	0.4796
Diet × Age	8	530	0.8927	3.87	0.0002
Sex × Age	8	530	0.9243	2.66	0.0072
Diet × ToD	8	530	0.9786	0.72	0.6729
Sex × ToD	8	530	0.9816	0.62	0.7624
Age × ToD	16	810	0.9693	0.52	0.9377

Results and Discussion

The frequency and the duration of bouts of different classes of activity in *B. tryoni* were affected by diet (sugar only, sugar + yeast), age (4, 10 or 30 days post-eclosion) and sex. There was, however, substantial variation in the effects across the treatment groups, with influential diet × age and age × sex interactions. MANOVA indicated that frequency of recorded behavioural states varied with the diet of *B. tryoni*, and the interactions of diet × age and age × sex (Fig. 1 and Table 1). Similarly, the total duration spent by *B. tryoni* performing each activity over the 10 min period assessed varied with a diet × age interaction (Fig. 2 and Table 2). Strong effects

of time of day on activity are reported for another tephritid fruit fly species (*Anastrepha ludens*; Zou *et al*., 2011), although such effects were absent from the present study (Tables 1 and 2), perhaps because fly activity was recorded outside the times of day when sexual advertisement, courtship and mating occur.

Diet and age

Diet had a significant effect on the frequency (Table 3) and duration (Table 4) of flight. The frequency and duration of flight tended to be higher in flies fed sugar + yeast

Fig. 2. Total duration of flying, walking, grooming and inactivity in relation to sex, age and diet in mass-reared *Bactrocera tryoni* over a period of 10 min. White bars represent sugar only diets; grey bars represent the sugar + yeast hydrolysate (YH) diet. Duration of (a) flight, (b) walking, (c) grooming and (d) inactivity in female flies. Duration of (e) flight, (f) walking, (g) grooming and (h) inactivity in male flies. Raw data are presented as the mean ± SE.

Table 2. Summary of multivariate analysis of variance on the effects of diet (sugar only, sugar + yeast hydrolysate), sex, age (4, 10 and 30 days after eclosion), and time of day (ToD; morning 10.00–12.00 h; early afternoon 12.00–14.00 h; late afternoon 14.00–16.00 h) on the duration of locomotor activities (flight, walking, grooming, inactivity) in mass-reared *Bactrocera tryoni* over a period of 10 min, as conducted on Box-Cox transformed data.

Source	d.f.$_{num}$	d.f.$_{den}$	Wilks' λ	F	P
Intercept	4	265	0.0328	1955.33	< 0.0001
Diet	4	265	0.9432	3.99	0.0037
Sex	4	265	0.9940	0.40	0.8108
Age	8	530	0.9639	1.23	0.2802
Time of day	8	530	0.9896	0.35	0.9468
Diet × Sex	4	265	0.9829	1.15	0.3324
Diet × Age	8	530	0.9181	2.89	0.0037
Sex × Age	8	530	0.9465	1.85	0.0665
Diet × ToD	8	530	0.9740	0.88	0.5336
Sex × ToD	8	530	0.9683	1.08	0.3786
Age × ToD	16	810	0.9560	0.75	0.7407

[Correction added on 19 June 2013, after first online publication: *P* value for Intercept was changed from 0.0000 to < 0.0001.]

by variation in the frequency of walking and inactivity (Table 3) and the duration of grooming and inactivity (Table 4), respectively. Flies fed sugar only increased walking frequency steadily from 4 days through to 30 days post-eclosion (Fig. 3a). By contrast, flies fed sugar + yeast walked more often than flies fed sugar only at 4 and 10 days of age but then declined in walking frequency at 30 days post-eclosion. These changes in walking frequency were not matched by significant changes in walking duration (Table 4), which could indicate an overall tendency for shorter bouts of walking in flies that walked more often.

The frequency (Fig. 3b) and duration (Fig. 4b) of inactivity was consistent across ages for flies fed sugar + yeast. By contrast, flies fed sugar only showed infrequent bouts of inactivity, spending little time inactive when 4 days post-eclosion but this increased markedly at 10 and 30 days post-eclosion. Although still reproductively immature, development of flies fed sugar + yeast is well under way at 4 days post-eclosion. The development of flies fed sugar only is greatly delayed in comparison, although some flies on this diet do eventually exhibit some gonadal development (Pérez-Staples *et al*., 2011; Weldon & Taylor, 2011) and sexual activity (Pérez-Staples *et al*., 2011) and so, to some extent, the increase in inactivity of these flies at 10 and 30 days post-eclosion might reflect delayed effects of development. However, as time passes, it is likely that flies suffered increasingly from deleterious effects of malnutrition and this might substantially explain the increased prevalence of inactivity. There is recent evidence of compensatory feeding in *B*. *tryoni* (Fanson *et al*., 2009; Fanson & Taylor, 2012a, 2012b). This offers a credible explanation for both low levels of inactivity in young flies (4 days post-eclosion) and increased activity in older (30 days post-eclosion) flies fed sugar only because flies search for food with different nutritional composition to obtain essential nutrients that are lacking in a sugar-only diet.

The duration of grooming activity was also consistent across ages for flies fed sugar + yeast, whereas flies fed sugar only

hydrolysate compared with flies fed sugar only (Fig. 1). Being an energetically demanding activity (Lehmann & Schützner, 2010), it is not surprising that flight activity tended to be greater in flies receiving sugar + yeast diets compared with flies receiving sugar only (Fig. 1). The effects of diet on flight activity were similar for males and females of all ages, at all times of day, and so the physiological effects of yeast supplementation on flight activity would appear to be common across these contexts.

Diet also influenced patterns of other measured activities but, in each case, the effect of diet varied with age. The significant diet × age interaction identified by MANOVA for both the frequency and duration of bouts of activity was driven

Table 3. Summary of univariate analysis of variance on the effects of diet (sugar only, sugar + yeast hydrolysate), sex, age (4, 10, 30 days after eclosion), and time of day (ToD; morning 10.00–12.00 h; early afternoon 12.00–14.00 h; late afternoon 14.00–16.00 h) on the frequency of locomotor activity in mass-reared *Bactrocera tryoni* over a period of 10 min, as conducted on Box-Cox transformed data.

	d.f.	Flight		Walking		Grooming		Inactivity	
		F	P	F	P	F	P	F	P
Intercept	1	265.92	< 0.0001	1439.65	< 0.0001	581.32	< 0.0001	894.28	< 0.0001
Diet	1	9.92	0.0018	9.03	0.0029	0.57	0.4491	0.19	0.6624
Sex	1	0.21	0.6489	0.42	0.5161	0.29	0.5880	0.09	0.7613
Age	2	0.08	0.9222	1.41	0.2466	0.42	0.6584	1.86	0.1574
ToD	2	0.07	0.9300	0.13	0.8822	0.45	0.6360	1.31	0.2724
Diet × Sex	1	0.20	0.6517	1.89	0.1705	2.57	0.1099	0.11	0.7400
Diet × Age	2	0.17	0.8463	4.22	0.0157	2.64	0.0732	8.91	0.0002
Sex × Age	2	5.15	0.0064	1.46	0.2341	3.62	0.0280	0.59	0.5561
Diet × ToD	2	0.42	0.6594	0.36	0.7016	1.68	0.1874	0.31	0.7341
Sex × ToD	2	0.92	0.3983	1.59	0.2065	0.15	0.8641	0.33	0.7219
Age × ToD	4	0.68	0.6069	0.45	0.7758	0.35	0.8412	0.55	0.6978
Error	268								
Total	287								

[Correction added on 19 June 2013, after first online publication: *P* value for Diet × Age was changed from 0.00018 to 0.0002.]

Table 4. Summary of univariate analysis of variance on the effects of diet (sugar only, sugar + yeast hydrolysate), sex, age (4, 10, 30 days after eclosion), and time of day (ToD; morning 10.00–12.00 h; early afternoon 12.00–14.00 h; late afternoon 14.00–16.00 h) on the duration of locomotor activities in mass-reared *Bactrocera tryoni* during a period of 10 min, as conducted on Box-Cox transformed data.

|	d.f.	Flight		Walking		Grooming		Inactivity	
		F	P	F	P	F	P	F	P
Intercept	1	214.56	< 0.0001	860.98	< 0.0001	567.61	< 0.0001	742.60	< 0.0001
Diet	1	11.65	0.0007	0.03	0.8666	0.35	0.5528	0.09	0.7614
Sex	1	0.52	0.4707	0.34	0.5612	0.37	0.5412	0.32	0.5748
Age	2	0.17	0.8439	1.07	0.3437	0.17	0.8415	3.32	0.0375
ToD	2	0.32	0.7287	0.16	0.8562	0.16	0.8500	0.64	0.5303
Diet × Sex	1	0.39	0.5333	0.09	0.7587	3.57	0.0600	0.60	0.4386
Diet × Age	2	0.30	0.7384	0.77	0.4639	3.89	0.0216	6.51	0.0017
Sex × Age	2	4.53	0.0116	0.31	0.7310	1.44	0.2386	0.35	0.7073
Diet × ToD	2	0.81	0.4467	1.33	0.2670	0.56	0.5701	0.84	0.4318
Sex × ToD	2	0.47	0.6276	3.06	0.0484	0.73	0.4807	0.37	0.6910
Age × ToD	4	1.00	0.4081	0.66	0.6180	0.88	0.4759	1.36	0.2466
Error	268								
Total	287								

[Correction added on 19 June 2013, after first publication: Inactivity *P* value for Diet × Age was changed from 0.00018 to 0.0002.]

tended to spend more time grooming when 4 days post-eclosion than when 10 or 30 days post-eclosion (Fig. 4a). The consistency in grooming activity of flies on the sugar + yeast diet at the different ages tested makes sense given the need for grooming is not expected to change with developmental stage. Patterns in flies fed sugar only may relate to physical rather than nutritional differences in the diet. Yeast hydrolysate is sticky, as are excreta of flies provided this diet, and so flies in cages containing this diet might be expected to maintain relatively high levels of grooming. The sugar-only diet, and the resulting excreta, is not nearly as sticky and this likely explains the lower levels of grooming at 10 and 30 days post-eclosion. However, this does not explain the elevated levels of grooming in flies fed sugar only at 4 days post-eclosion. For flies on a sugar-only diet, the main source of nutrition other than sugar would be the sticky excreta of other flies. In the

days immediately after eclosion, this excreta may contain some nutrients carried over from the pupal stage (Nestel *et al.*, 2004). Although such nutrients might still be reasonably abundant at 4 days post-eclosion, they are likely depleted by 10 days. Hence, the elevated levels of grooming in the sugar-fed flies at 4 days post-eclosion might be a response to the presence of more nutritious and sticky excreta at this time and elevated rates of coprophagy.
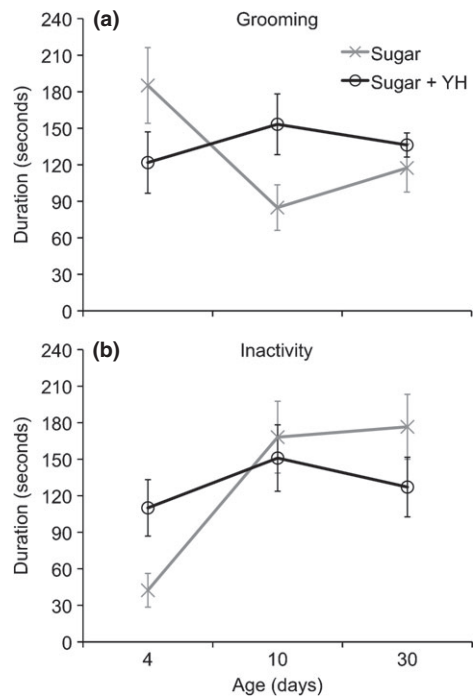
Sex and age

The significant sex × age interaction on overall frequency of activities identified by MANOVA (Table 1) was driven by variation in frequency of flight and grooming (Fig. 5 and Table 3). Sex differences in the frequency of flight activity
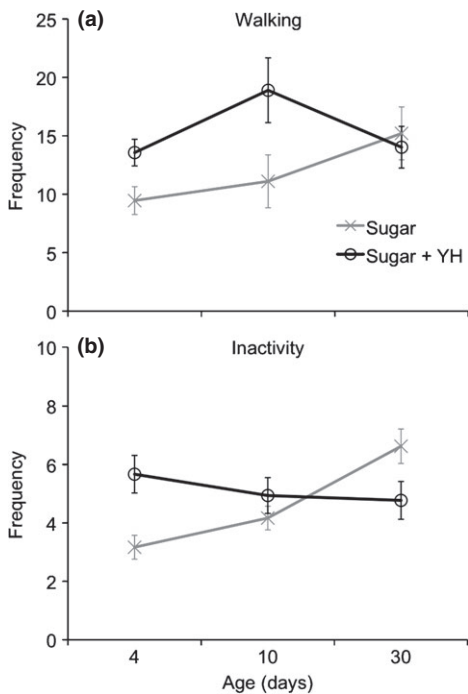
Fig. 3. Variation in the frequency of (a) walking and (b) inactivity of fertile mass-reared *Bactrocera tryoni* over a period of 10 min in response to diet and age. The diets provided to the flies comprised sugar and yeast hydrolysate (YH) or sugar only. Raw data are presented as the mean ± SE.


Fig. 4. Variation in the duration of (a) grooming and (b) inactivity of fertile mass-reared *Bactrocera tryoni* over a period of 10 min in relation to diet [sugar, sugar + yeast hydrolysate (YH)] and age. Raw data are presented as the mean ± SE.

were evident only at the oldest age tested. Both sexes showed a pattern of increased flight activity from 4 to 10 days post-eclosion but, whereas females then continued increasing flight activity levels to day 30, males instead exhibited a substantial decline by day 30 (Fig. 5). Given that there was no evidence of differences between diet groups, we cannot ascribe this age effect to either maturation or ageing, both of which are linked closely to diet (Carey *et al*., 2008; Lee *et al*., 2008; Maklakov *et al*., 2008; Fanson *et al*., 2009; Fanson & Taylor, 2012b).

Consistent with the previous study of Weldon *et al*. (2010), no sex differences in patterns of walking and inactivity of sexually immature *B. tryoni* were found (4 days post-eclosion; Tables 3 and 4). At this ontological stage, both sexes must forage for dietary resources needed for sexual development (Pérez-Staples *et al*., 2007); females require hydrolysed yeast in their diets to produce eggs and males also need it to reach peak sexual performance (Pérez-Staples *et al*., 2011). With similar selection pressures acting on females and males at this life stage, similar behavioural responses can be expected. Weldon *et al*. (2010) propose that high levels of grooming in mass-reared *B. tryoni* relative to their wild counterparts may reflect conditions associated with high population density. An increased prevalence of pathogens (Anderson & May, 1981) could lead to selection for increased investment in activities related to resistance (Hochberg, 1991; Reeson *et al*., 1998). Considering the 4 day-old flies that fed sugar + yeast, a close match to the flies used in the study by Weldon *et al*. (2010), we observe low levels of grooming compared with the frequency

and duration of walking, which is akin to that of wild flies reported by Weldon *et al*. (2010) for a similar age and the same diet. It is possible that variation in developmental conditions at the mass-rearing facility makes a considerable contribution to the behaviour of *B. tryoni*; many studies using the same mass-reared strain report substantial variation between cohorts in adult eclosion, flight ability, sex ratio and mortality under stress (Dominiak *et al*., 2002, 2008; Collins *et al*., 2008), and aspects of mating behaviour (Pérez-Staples *et al*., 2007; Prabhu *et al*., 2008; Pérez-Staples *et al*., 2008).

Inactivity and quality control

Weldon *et al*. (2010) suggest that data on the frequency and duration of inactivity offers a simple but potentially sensitive and biologically relevant test of quality for mass-reared tephritids. Standardized quality control tests measuring the performance of mass reared tephritid flies are available, some of which assess locomotor performance, notably flight ability (FAO/IAEA/USD, 2003), startle (Boller *et al*., 1981) and predator avoidance (Hendrichs *et al*., 2007). Activity is generally related to health and vigour in insects (Martin, 2003) and is an important criterion used to assess sterile male performance (Hendrichs *et al*., 2007; Collins *et al*., 2008). The assessment of general activity lends itself readily to automation, using activity monitors of the type adopted for studies of *Drosophila* (Fernández *et al*., 1999; Martin
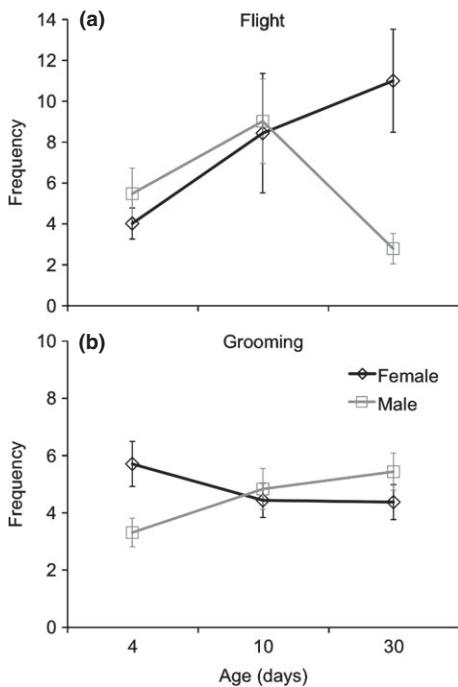
Fig. 5. Variation in the frequency of (a) flight and (b) grooming by fertile mass-reared *Bactrocera tryoni* over a period of 10 min in response to fly sex and age. Raw data are presented as the mean ± SE.

et al., 1999; Martin, 2003) to quantify bouts of activity and inactivity. Video-recordings, and especially automated systems, would mitigate the inter-individual and session variation in experimenter skills and observer effects that can easily affect startle and predator avoidance methods (Martin & Bateson, 2007) and thus provide an effective, straighforward, highly repeatable alternative quality control measure in mass-rearing environments. This method might also be useful for providing an activity threshold for the selection of flies for control release. Weldon *et al*. (2010) report that activity levels of mass-reared domesticated flies are diminished relative to wild type flies, and that even further reductions are induced by irradiation for the induction of reproductive sterility. If the activity patterns of wild flies provide a quality benchmark defined by adaptation for field conditions, then we can interpret the lower activity seen in domesticated and irradiated flies as indicating diminished quality (i.e. fitness for field conditions). More active mass-reared flies would be more similar to wild flies and, by this standard, could be interpreted as being of higher quality than less active mass-reared flies. Further research is required to derive appropriate activity benchmarks if activity assays are to be developed for the assessment of fly quality in mass-rearing programmes.

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