A Review of the Scientific Literature for Optimal Conditions for Mass Rearing *Tenebrio molitor* (Coleoptera: Tenebrionidae)[1]

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Abstract Several insect species are mass produced for animal feeds, fish bait, or human consumption. The yellow mealworm, *Tenebrio molitor* L. (Coleoptera: Tenebrionidae), is one of the more-popular insects considered for this use. To make *T. molitor* competitive with more-traditional protein sources, mass production must be optimized. Herein, we review the scientific literature on the range and optimal environmental, physical, and dietary conditions for the rearing *T. molitor*. Our review identified that: (a) the optimum temperature for *T. molitor* growth is 25–28°C; (b) *T. molitor* larval growth rate is greatest at $\geq$70% relative humidity (RH) with an optimum range between 60–75% RH, and; (c) optimal growth is achieved with diets containing 5–10% yeast, 80–85% carbohydrate, and the addition of B-complexvitamins.

Key Words *Tenebrio molitor*, temperature, relative humidity, diet, rearing

The rapid growth of the world population has forced consideration of nontraditional food sources to achieve food security, preserve natural resources, and provide for sustainable development. Since van Huis et al. (2013) promoted mass production of insects for food and feed, there has been a growing movement worldwide, and several "start-up" companies have been established for mass production of insects as an alternative source of protein.

Only a few insect species have been commercially mass produced for a protein source, either in an industrial or a "cottage" setup. These have been the black soldier fly, *Hermetia illucens* (L.) (Diptera: Stratiomyidae), the common housefly, *Musca domestica* L. (Diptera: Muscidae), silkworms, *Bombyx mori* L. (Lepidoptera: Bombycidae), several species of termites (Isoptera), several species of grasshoppers and locusts (Orthoptera), and the yellow mealworm, *Tenebrio molitor* L. (Coleoptera: Tenebrionidae).

Tenebrio molitor is relatively easy to culture. Small producers have been rearing it in Europe and elsewhere, either for animal feed, fish bait, or human consumption. They generally culture the insects in trays with a substrate where the different stages develop separately. This process is highly dependent on manual labor, as the feeding remains and waste materials must be separated from the insects by

sieving the contents of the trays. Although the process can be mechanized (Cortes Ortiz et al. 2016; Oonincx and de Boer 2012), the use of insects for feed or food will only be a reality if production costs are competitive when compared to production of other, more-traditional protein sources (e.g., soybean, fishmeal). Optimization of production costs may be achieved by using low-cost food sources, automated rearing techniques, and optimal growth conditions.

To our knowledge, no single paper contains all the information related to the optimal conditions for the rearing *T. molitor*. Our objective here was to review and compile the studies identifying the range and optimal environmental, physical, and dietary conditions for rearing *T. molitor*. We have divided our review of the pertinent literature into four areas of: (1) biology and life cycle; (2) environmental conditions; (3) diet components, and; (4) nutritive value and use.

Biology and Life Cycle

Tenebrio molitor feeds primarily on farinaceous materials such as flour and meal. It is considered a pest where these are produced and stored. Females produce an average of 250–500 eggs which they oviposit singly or in small clusters attached to the substrate or the walls and floor of the containers in which they are oviposited (Cotton 1927; Ghaly and Alkoaik 2009; Hardouin and Mahoux 2003; Hill 2002; Manojlovic 1987; Spencer and Spencer 2006;). Eggs hatch in 4 d at 26–30°C (Cotton 1927; Siemianowska et al. 2013) but may take up to 34 d to hatch at 15°C (Kim et al. 2015). Duration of the larval stage varies from 57 d in controlled conditions (Weaver and McFarlane 1990) to 629 d in ambient natural conditions (Cotton 1927), with an average duration of 112–203.3 d (Damborsky et al. 2000; Martin et al. 1976). Larvae undergo several molts, varying from a minimum of 9 (Cotton 1927; Hill 2002) to a maximum of 23 (Ludwig 1956), with an average of 11–19 instars (Damborsky et al. 2000; Ludwig 1956). The number and the duration of the larval instars obviously influences the total duration of the life cycle of *T. molitor*.

As larvae approach pupation, they enter a short period of latency in which they appear as a "C" shape before turning into a pupa. The pupal stage lasts 6 (Cotton 1927; Ghaly and Alkoaik 2009) to 20 d (Hill 2002). Adults first emerge as whitish-colored beetles with soft exoskeletons, which gradually harden and darken. Mating and oviposition begin in about 3 d after emergence (Manojlovic 1987), and the adult stage lasts 16 to 173 d (Damborsky et al. 2000) with an average of 31.8 (Urs and Hopkins 1973a) to 62 d (Damborsky et al. 2000). The entire life cycle occurs in the same habitat or vicinity and, with optimal conditions, lasts 75 (Spencer and Spencer 2006) to 90 d (Hardouin and Mahoux 2003) (Table 1).

Environmental Conditions

Temperature and relative humidity (RH). Temperatures commonly used to rear mealworms are 25–28°C (Kim et al. 2015; Koo et al. 2013; Ludwig 1956; Punzo 1975; Punzo and Mutchmor 1980; Spencer and Spencer 2006,), with an absolute minimum of 10°C (Punzo and Mutchmor 1980) and a maximum of 35°C (Martin et al. 1976; Punzo and Mutchmor 1980). Temperatures <17°C inhibit embryonic development (Koo et al. 2013) and temperatures >30°C increase death rates (Koo

Table 1. Biological and life cycle parameters of *Tenebrio molitor*. Minimum, maximum, and average values for the number of eggs, length, lifespan, duration of the life cycle, and number of larval instars. A dash (—) indicates no data.

	Minimum	Average	Maximum
Number of eggs	77 (Cotton 1927) 250 (Ghaly and Alkoaik 2009)	250 (Spencer and Spencer 2006) 276 (Cotton 1927) 280 (Hardouin and Mahoux 2003) 414 (Manojlovic 1987) 400–500 (Ghaly and Alkoaik 2009)	500 (Hill 2002) 576 (Cotton 1927) 1,000 (Ghaly and Alkoaik 2009)
Length (mm)			
Larvae	20 (Ghaly and Alkoaik 2009) 28 (Hardouin and Mahoux 2003)	16 (Barker et al. 1998) 28 (Hill 2002)	25 (Ghaly and Alkoaik 2009) 31.6 (Park et al. 2014) 32 (Hardouin and Mahoux 2003)
Adult	12 (Hill 2002) 15.5 (Kim et al. 2015)	—	16 (Hill 2002; Kim et al. 2015)
Body mass (mg)			
Larvae	75 (Martin et al. 1976) 130 (Makkar et al. 2014) 140 (Tschinkel and Willson 1971)	111 (Martin et al. 1976) 120 (Connat et al. 1991) 126 (Finke 2002) 191 220 (Kim et al. 2016)	134 (Ramos-Elorduy et al. 2002) 145 (Martin et al. 1976) 160 (Makkar et al. 2014) 182.7 (Ghaly and Alkoaik 2009) 190 (Tschinkel and Willson 1971) 220 (Kim et al. 2016)
Adult	—	136 (Finke 2002)	—

Table 1. Continued.

	Minimum	Average	Maximum
Duration (d)			
Lifespan (complete cycle)	75 (Spencer and Spencer 2006) 90 (Hardouin and Mahoux 2003) 181 (Urs and Hopkins 1973a) 280 (Hill 2002)	80–83.7 (Park et al. 2012) 189 (Urs and Hopkins 1973a) 294 (Damborsky et al. 2000)	90 (Spencer and Spencer 2006) 120 (Hardouin and Mahoux 2003) 196 (Urs and Hopkins 1973a) 630 (Hill 2002)
Egg stage	4 at 26–31°C (Cotton 1927) 4–6 (Siemianowska et al. 2013) 5 at 35°C (Kim et al. 2015) 7 (Damborsky et al. 2000) 10 (Hill 2002)	7 (Hardouin and Mahoux 2003; Li et al. 2016b) 7.55 at 25°C (Manojlovic 1987) 7–8 (Park et al. 2014) 9.2 (Damborsky et al. 2000) 12.6 at 20°C (Manojlovic 1987) 15 (Ghaly and Alkoaik 2009)	10 (Spencer and Spencer 2006) 12 (Hill 2002) 15 (Damborsky et al. 2000) 19 at 18–21°C (Cotton 1927) 34 at 15°C (Kim et al. 2015)
Larval stage	57 (Weaver and McFarlane 1990) 87.7 (Urs and Hopkins 1973a) 110.8 at 30°C (Koo et al. 2013) 120 (Hill 2002) 127 (Kim et al. 2015) 129 (Morales-Ramos et al. 2015) 143 (Morales-Ramos et al. 2010) 180 (Ghaly and Alkoaik 2009) 280 in nature (Cotton 1927)	112 (Martin et al. 1976) 120 at 25°C (Ludwig 1956) 151.15 (Urs and Hopkins 1973a) 173 at 30°C (Ludwig 1956) 180 (Cotton 1927) 203.3 (Damborsky et al. 2000) 241.9 (Park et al. 2014)	202.5 (Urs and Hopkins 1973a) 216 (Kim et al. 2015; Morales-Ramos et al. 2015) 240 (Ghaly and Alkoaik 2009) 244 at 17°C (Koo et al. 2013) 540 (Hill 2002) 629 in nature (Cotton 1927)

Table 1. Continued.

	Minimum	Average	Maximum
Pupal stage	6 at 27°C (Cotton 1927) 6 at 28°C (Ghaly and Alkoaik 2009) 6.51 at 27.5°C (Kim et al. 2015) 7.7 (Urs and Hopkins 1973a)	5–6 (Siemianowska et al. 2013) 7 (Hardouin and Mahoux 2003) 7.3 (Morales-Ramos et al. 2010) 7.6 (Damborsky et al. 2000) 8.9 (Urs and Hopkins 1973a) 9 (Hill 2002)	10.1 (Urs and Hopkins 1973a) 18 at 18°C (Cotton 1927; Ghaly and Alkoaik 2009) 20 (Hill 2002; Kim et al. 2015)
Adult	16 (Damborsky et al. 2000) 25 (Urs and Hopkins 1973a) 30 (Hill 2002) 37 (Ghaly and Alkoaik 2009) 38 (Cotton 1927)	31.8 (Urs and Hopkins 1973a) 61.5 (Cotton 1927) 62.05 (Damborsky et al. 2000)	60 (Hill 2002) 94 (Cotton 1927) 96 (Ghaly and Alkoaik 2009) 105 (Urs and Hopkins 1973a) 173 (Damborsky et al. 2000)
Number of larval instars	9 (Cotton 1927; Hill 2002) 11 at 25°C (Ludwig 1956) 12.2 (Urs and Hopkins 1973a) 14 (Morales-Ramos et al. 2015) 15 at 30°C (Ludwig 1956)	11 (Damborsky et al. 2000) 13 (Koo et al. 2013) 13.2 at 25°C (Ludwig 1956) 14 (Morales-Ramos et al. 2010) 14.15 (Urs and Hopkins 1973a) 15 (Hardouin and Mahoux 2003) 15–17 (Park et al. 2014) 17–19 (Cotton 1927) 19.1 at 30°C (Ludwig 1956)	15 at 25°C (Ludwig 1956) 15 (Morales-Ramos et al. 2010) 16.1 (Urs and Hopkins 1973a) 18 (Morales-Ramos et al. 2015) 20 (Cotton 1927; Hill 2002; Park et al. 2014) 23 at 30°C (Ludwig 1956)

Table 2. Minimum, maximum, and optimal temperature (°C) conditions used on the rearing of eggs, larvae, pupae, and adults of mealworms.

	Minimum	Optimal	Maximum
Eggs	10 (Punzo and Mutchmor 1980) 15 (Kim et al. 2015) 17 (Koo et al. 2013)	23–27 (Manojlovic 1987) 25 (Murray 1960, 1968; Punzo and Mutchmor 1980) 25–27 (Siemianowska et al. 2013)	30 (Manojlovic 1987) 35 (Kim et al. 2015; Punzo and Mutchmor 1980)
Larvae	10 (Punzo and Mutchmor 1980) 17 (Koo et al. 2013) 20 (Martin et al. 1976)	25 (Ludwig 1956; Murray 1960, 1968; Punzo 1975; Punzo and Mutchmor 1980) 27–28 (Kim et al. 2015; Koo et al. 2013; Spencer and Spencer 2006)	30 (Koo et al. 2013; Ludwig 1956) 35 (Martin et al. 1976; Punzo and Mutchmor 1980)
Pupae	10 (Punzo and Mutchmor 1980) 18 (Cotton 1927) 21 (Hardouin and Mahoux 2003)	25 (Murray 1960, 1968; Punzo 1975) 27 (Cotton 1927) 27.5 (Kim et al. 2015) 28 (Ghaly and Alkoaik 2009) 27–33 (Hardouin and Mahoux 2003)	35 (Punzo and Mutchmor 1980)
Adult	10 (Punzo and Mutchmor 1980) 14–16 (Dick 1937)	25 (Murray 1960, 1968; Punzo 1975)	35 (Punzo and Mutchmor 1980)

et al. 2013; Ludwig 1956). The minimal and maximum lethal temperatures for exposure periods of 24 h are 7–8°C (Mutchmor and Richards 1961) and 40–44°C (Altman and Dittmer 1973; Martin et al. 1976), respectively. There are no significant differences in the temperature requirements for the different stages of development of this species (Table 2).

Mealworm response to RH varies, although their resilience depends largely on temperature (Table 3). In all studies focused on the influence of RH on the development of *T. molitor*, optimum values varied from 60% (Manojlovic 1987) to 75% RH (Punzo 1975; Punzo and Mutchmor 1980).

Temperature combined with RH affects both the number and the length of the instars and also influences the water absorption capacity of the different stages of the life cycle. The pupal stage is the most resilient to extreme conditions of

Table 3. Minimum, maximum, and optimal relative humidity (%) conditions on the rearing of eggs, larvae, pupae, and adults of mealworms.

	Minimum	Optimal	Maximum
Eggs	12 (Punzo 1975; Punzo and Mutchmor 1980)	60–75 (Manojlovic 1987) 70 (Murray 1960, 1968) 75 (Punzo and Mutchmor 1980)	98 (Punzo 1975; Punzo and Mutchmor 1980)
Larvae	12 (Punzo 1975; Punzo and Mutchmor 1980) 30 (Fraenkel 1950)	75 (Punzo 1975; Punzo and Mutchmor 1980) 60–70 (Spencer and Spencer 2006) 70 (Fraenkel 1950; Fraenkel and Blewett 1944; Hardouin and Mahoux 2003; Martin et al. 1976; Murray 1960, 1968)	98 (Punzo 1975; Punzo and Mutchmor 1980)
Pupae	12 (Punzo 1975; Punzo and Mutchmor 1980)	70 (Murray 1960, 1968) 75 (Punzo 1975; Punzo and Mutchmor 1980)	98 (Punzo 1975; Punzo and Mutchmor 1980)
Adult	12 (Punzo 1975; Punzo and Mutchmor 1980) 20 (Dick 1937; Hardouin and Mahoux 2003)	70 (Murray 1960, 1968) 75 (Punzo 1975; Punzo and Mutchmor 1980) 90–100 (Hardouin and Mahoux 2003)	98 (Punzo 1975; Punzo and Mutchmor 1980)

temperature and RH while the egg and early larval stages are the most sensitive (Punzo and Mutchmor 1980).

Oviposition does not occur at temperatures below 14°C with an RH of 65%, but even at the optimal temperature of 27°C in low humidity (20%), oviposition is significantly reduced due to the loss of body moisture by evaporation and subsequent death (Dick 1937). Female adult activity is also reported to be greatest at 90–100% RH (Hardouin and Mahoux 2003).

At 10°C (Punzo 1975; Punzo and Mutchmor 1980) and 12.5°C (Kim et al. 2015), water absorption in eggs is reduced and embryological development halts. Whatever the temperature, extremely dry conditions of about 12% RH potentiate water losses from the eggs, culminating in death due to desiccation (Punzo 1975).

The growth rate of *T. molitor* larvae is highly dependent on moisture, with higher growth rates at 90–100% RH (Hardouin and Mahoux 2003) and even at 70% RH, but the rate slows at 30% RH and hardly occurs at 13% (Fraenkel 1950). During times of extremely dry conditions, mealworm larvae may cease feeding and become inactive until RH rises to more-favorable levels (Urs and Hopkins 1973). In

Table 4. Physical conditions and population density used on the rearing of mealworms.

Population Density (Larvae/dm^2)	Tray Dimensions (cm)	Area (dm^2)	Volume (dm^3)	References
8.4				Morales-Ramos et al. 2012
50–150	11 × 18 × 3	1.98	0.6	Connat et al. 1991*
80–145	16 × 21.5 × 7	3.44	2.4	Park et al. 2012
48	48 × 49.5 × 10.5	23.76	25	Park et al. 2014
2	30 × 15	4.50		Spang 2013
30	17.5 × 9.3 × 6.3	1.63	1	Oonincx et al. 2015
30	40 × 30 × 25	12	30	Alves et al. 2016
40–60	40 × 17 × 11	6.80	7.5	Rho and Lee 2016
30	10 (diameter) × 28	0.79	2.2	Dick 1937
3	60 × 45 × 15	27	40.5	Ghaly and Alkoaik 2009
375–750		6		Martin et al. 1976
1–20	10 (diameter)	0.79		Tschinkel and Willson 1971

* The population density was the object of study.

general, the higher the RH, the higher the growth rate, but high levels of moisture favor the growth colony contaminants (e.g., fungi, other microorganisms, mites) (Fraenkel 1950).

Larval instar stages are shorter, and the numbers of instars are greater at 30°C than at 25°C, resulting in a longer total larval developmental period at 30° than at 25°C (Ludwig 1956). Temperatures below 10°C and above 35°C clearly constitute stress conditions for this species, although the minimal lethal temperature is 7°C and the maximum lethal temperature is 44°C (Punzo and Mutchmor 1980).

Tenebrio molitor development and survival are affected by the interaction of temperature and moisture. The effect of temperature is potentiated at extreme moisture conditions, and the effect of moisture is also more critical at extreme temperatures (Punzo and Mutchmor 1980). For example, at optimal temperatures of 25–27.5°C, *T. molitor* displays no stress even under extreme humidity conditions and long exposure periods. Moreover, while at 25°C the decrease in moisture has no significant effects on adults, larvae, or pupae, a temperature of 10°C increases mortality (Punzo and Mutchmor 1980).

Population density. Our search found that several densities of larvae have been used in experiments, depending on the objective of the study. These have ranged from 1 larva/dm^2 (i.e., a single larva in a 10-cm diameter Petri dish) to a maximum of 750 larva/dm^2 (Table 4). Morales-Ramos et al. (2012) studied the effect of density on adult population survival and production, obtaining an optimal density of 8.4 adults/dm^2 for mass production purposes.

Population density influences the number and the duration of the larval molts, with higher population densities resulting in fewer larval instars and smaller-sized penultimate larvae (Connat et al. 1991; Morales-Ramos et al. 2012; Morales-Ramos and Rojas 2015; Tschinkel and Willson 1971; Weaver and McFarlane 1990). Early studies have noted inhibition of pupation and cannibalism associated with crowding in tenebrionid beetles (Tschinkel and Willson 1971) as well as the occurrence of incomplete transformations and slower growth rates due to a reduced feeding opportunity induced by conspecific competition (Weaver and McFarlane 1990). Although high population density leads to a reduction in food consumption, the lower growth rates are also a consequence of the decrease in the efficiencies of both ingested and digested food conversion (Morales-Ramos and Rojas 2015). Overcrowding also reduces female progeny and reproductive output, with rearing of two insects per chamber being the most productive in terms of number of progeny produced per female (Morales-Ramos et al. 2012). Furthermore, overpopulation also affects the prepupal stage, with isolated larvae exhibiting accelerated larval–pupal apolyses and a reduced number of larval instars (Connat et al. 1991). Moreover, the metabolism of larvae in overcrowded populations results in a substantial increase in temperature that can easily be lethal.

Photoperiod. Mealworms are negatively phototropic and phototactic (Balfour and Carmichael 1928; Cloudsley-Thompson 1953), with adults and larger larvae positioning below the surface of the substrate during daylight and coming to surface in darkness. Although photoperiod influences growth and development of mealworms (Tyshchenko and Ba 1986), the response to photoperiod tends to disappear under constant conditions, when *T. molitor* becomes arrhythmic (Cloudsley-Thompson 1953). Recent studies show that larval development was optimal in long-day conditions, with lower development times observed in photoperiods of 14L:10D (Kim et al. 2015). Photoperiod also influences eclosion rates, with higher rates in long-day conditions, i.e., 45.5% at 14L:10D versus 24.2% at 10L:14D (Kim et al. 2015) and pupation with 100% pupation induced by a 12L:12D regime at 25°C. Under other photoperiod regimes, long-day result in more-protracted delays than did shorter-day conditions. However, at 30°C the photoperiodic response was reversed, with an inhibition of pupation under 12L:12D and enhancement under 18L:6D (Tyshchenko and Ba 1986) (Table 5).

Oxygen. Low oxygen concentrations increase larval mortality and hypertrophy of tracheae and other respiratory exchange organs (Greenberg and Ar 1996; Loudon 1988, 1989), and hypoxia conditions (10–10.5% O_2) inhibit growth rate, lead to the development of abnormalities, and to a higher proportion of females in mealworm populations (Loudon 1988). Although development time is similar for both normoxia (21% O_2) and hyperoxia (40% O_2) conditions, hyperoxia induces a lower number of instars, thus resulting in lower final larval biomass than with normoxia (Greenberg and Ar 1996).

Dietary Components and Conditions

Mealworms have the ability to select foods in order to balance their diet ratio/intake according to their nutritional needs (Morales-Ramos et al. 2011; Rho and Lee 2014, 2016; Urrejola et al. 2011). *Tenebrio molitor* can be fed only with bran, which

Table 5. Photoperiod regimes used in the rearing of *Tenebrio molitor*.

Light:Dark (h)	References
12:12	Allen et al. 2012; Gerber and Sabourin 1984; Greenberg and Ar 1996; Kim et al. 2015; Lardies et al. 2014; Lv et al. 2014; Martin et al. 1976; Menezes et al. 2014; Oonincx et al. 2015; Rho and Lee 2014, 2016; Tang et al. 2012; Urrejola et al. 2011; van Broekhoven et al. 2015
Darkness	Morales-Ramos et al. 2013; Oonincx et al. 2010; Punzo 1975; Punzo and Mutchmor 1980
14:10	Kim et al. 2015; Koo et al. 2013; Morales-Ramos et al. 2010; Ravzanaadii et al. 2012
10:14	Alves et al. 2016; Kim et al. 2015; Rho and Lee 2016; Weaver and McFarlane 1990
16:8	Tindwa et al. 2015; Urs and Hopkins 1973a

contains all the necessary nutrients but not in optimal proportions, making diet supplementation beneficial (Morales-Ramos et al. 2010) at different developmental stages. A diet composed of bran and a water source (e.g., fresh vegetables such as carrot, apple, potato, or cabbage) and/or a protein source (e.g., beer yeast, casein, or soy protein) is the most common diet composition in mealworm industry and laboratory rearing facilities (Table 6).

Nutrient intake plays a central role in *T. molitor* life cycle, influencing development time (Davis 1970a; Fraenkel 1950; Martin and Hare 1942; Morales-Ramos et al. 2010, 2013; Rho and Lee 2016; Urrejola et al. 2011; van Broekhoven et al. 2015), fertility (Gerber and Sabourin 1984; Manojlovic 1987; Morales-Ramos et al. 2013; Urrejola et al. 2011), number of instars (Morales-Ramos et al. 2010), survival rate (Morales-Ramos et al. 2010; van Broekhoven et al. 2015), the intensity and period of oviposition (Manojlovic 1987; Morales-Ramos et al. 2013), and progeny production by increasing the number of eggs and decreasing adult mortality (Gerber and Sabourin 1984).

Water intake. Although *T. molitor* can survive under extremely dry conditions for extended periods of time, being able to obtain water from both the food ingested (even substances with low water content) and the atmosphere (Fraenkel and Blewett 1944), larvae grow faster in humid conditions higher than 70% RH. However, the development of mites, fungi, and other microorganisms enhanced by these high-humidity levels are undesirable to the mass rearing of this insect. Mealworms reared on dry substrates exhibit higher growth rates when there is any source of water (Mellandby and French 1958; Oonincx et al. 2015; Urs and Hopkins 1973b). In fact, on low-moisture substrates when metabolic water per unit of food is low (24–35 g water/100 g food), development may halt if there is no drinking water. When suffering from water deprivation, *T. molitor* larvae ingest less food, and the conversion rate of ingested food into body mass decreases (Murray 1968). The

Table 6. Diets and protein sources used on the rearing of mealworm.

Diet	Reference
Bran/flour + water source (vegetable or water)	Allen et al. 2012; Baek et al. 2015; Dick 2008; Greenberg and Ar 1996; Houbraken et al. 2016; Li et al. 2015; Ludwig 1956; Lv et al. 2014; Damborsky et al. 2000; Morales-Ramos et al. 2012; Punzo 1975; Ravzanaadii et al. 2012; Siemianowska et al. 2013; Spang 2013; Tschinkel and Willson 1971; Urs and Hopkins 1973b; Vinokurov et al. 2006; Xuegui et al. 2010;
Bran/flour + water source (vegetable or water) + protein source	Connat et al. 1991; Ghaly and Alkoaik 2009; Kim et al. 2015; Lardies et al. 2014; Li et al. 2016a; Manojlovic 1987; Murray 1968; Oonincx et al. 2010; Ramos-Elorduy et al. 1997; Tang et al. 2012; Weaver and McFarlane 1990
Varied artificial diet	Alves et al. 2016; Davis 1970a, 1974, 1978; Davis and Sosulski 1973a; Fraenkel 1950; Gerber 1975; Hardouin and Mahoux 2003; John et al. 1978; Martin and Hare 1941; Martin et al. 1976; Menezes et al. 2014; Morales-Ramos et al. 2010, 2013; Murray 1960; Oonincx et al. 2015; Rho and Lee 2014, 2015; Tindwa et al. 2015; Urrejola et al. 2011; van Broekhoven et al. 2016
Protein source	
Beer yeast	Gerber 1975; Ghaly and Alkoaik 2009; John et al. 1978; Lardies et al. 2014; Oonincx et al. 2010, 2015; Ramos-Elorduy et al. 1997; Tang et al. 2012; Tindwa et al. 2015; Urrejola et al. 2011; van Broekhoven et al. 2015; Weaver and McFarlane 1990
Casein	Davis 1970b, 1978; Davis and Leclercq 1969; Fraenkel 1950; Murray 1960; Rho and Lee 2014
Dried yeast	Connat et al. 1991; Murray 1960, 1968
Albumin	Morales-Ramos et al. 2010, 2013; Murray 1960; Rho and Lee 2014

Table 6. Continued.

Diet	Reference
Soy	Davis and Sosulski 1973b; Hardouin and Mahoux 2003; Manojlovic 1987; Morales-Ramos et al. 2013
Dry potato	Morales-Ramos et al. 2016
Lactalbumin	Davis 1970b; Davis and Leclercq 1969
Lactalbumin hydrolisate	Davis 1970b; Davis and Leclercq 1969
Aminoacid mixture	Davis 1974; John et al. 1978
Bird feed	Menezes et al. 2014
Bocaiuva (*Acrocomia aculeata*)	Alves et al. 2016
Cookie crumbs	Oonincx et al. 2015; van Broekhoven et al. 2015
Beef (blood, muscle, liver)	Martin and Hare 1941

addition of a source of water, such as carrots, results in the increase of survival rates ($\geq$80%) and reduces the development time from 145–151 d to 91–95 d (Oonincx et al. 2015).

The reported effects of water intake on biomass composition are contradictory. Urs and Hopkins (1973) observed that the availability of water increases the concentrations of total lipids while Oonincx et al. (2015) reported that the supplementation of the diets with a water source increases the moisture content but not the total fatty acids content. Both studies found no influence of water on mealworm fatty acid profile.

Protein. Dietary protein and amino acids greatly influence *T. molitor* life cycle, with direct benefits for larval development time, survival, and weight gain. Supplementation of diets with protein, even in small percentages, has been shown to increase the developmental rate of larvae. Diets supplemented with high protein content (33–39% dry mass) reduce the time for pupation from 191–227 d to 116–144 d at 28°C and 70% RH (Oonincx et al. 2015) and the time to 50% pupation from 95–168 to 79–95 d at 28°C and 65% RH (van Broekhoven et al. 2015), when compared with diets supplemented with low protein content (12% dry mass). Likewise, supplementation with protein increases survival rates from 84–88% to 88–92% (van Broekhoven et al. 2015) and from 19–52% to 67–79% (Oonincx et al. 2015).

The growth rate is enhanced by protein, with a weight gain per larva of 2.3–2.9 mg on a protein-free diet to 45.5–55.6 mg on a diet supplemented with 10 parts of yeast and 90 parts of whole ground wheat during a period of 4 wk (John et al. 1979). Supplementation with casein enhances growth rates from 4.08 g/g of larva with 3% casein to 6.16 g/g of larva with 20% casein (Davis 1970a). The benefits of adding protein to the diet are visible in pupae, with weight gains enhanced from 123 mg/g of

food with no supplement to 238 mg/g of food supplemented with protein (Morales-Ramos et al. 2013) and pupal weight from 117–145 mg with a low-protein diet (5% yeast) to 146–161 mg with a rich-protein (40% yeast) diet (van Broekhoven et al. 2015).

Fertility is also highly influenced by the presence of protein in mealworm diet, with increases in average female fecundity from 3 eggs/d in diets without protein increased to 6–7 eggs/d in diets supplemented with protein (Morales-Ramos et al. 2013). Urrejola et al. (2011) also obtained an increase in female fecundity from 5 to 12 eggs/d in diets with yeast at 20% when compared to diets with 2% yeast addition (w/w). Additionally, the period of maximum oviposition occurs earlier in females reared with soybean flour (9th–12th day) than in females reared only with wheat (12th–15th day).

Mealworms seemingly have a highly constrained body protein content with 2–3-fold increases in crude protein content (11.9–39.1% dry mass) of diets resulting in similar body protein composition (van Broekhoven et al. 2015). Yeast, at concentrations of 5–10%, is considered the best protein source acting as a feeding stimulant (Fraenkel 1950; Martin and Hare 1942). Other protein sources that provide optimal effects are 2–32% casein (Davis and Leclercq 1969) and at a lower level, lactalbumin (Davis and Leclercq 1969; Fraenkel 1950; Leclercq 1948). Zein, gliadin, and enzymatic hydrolysates from casein or lactalbumin have residual effects when compared to casein and lactalbumin (Fraenkel 1950). Although soybean is a rich protein source, it contains a potent trypsin inhibitor that negatively influences the larval growth (Birk et al. 1962).

The amino acid contents in the mealworm larval tissues are 8.9–9.9% alanine, 7.9–8.7% aspartic acid, 7.7–8.0% leucine, 6.5–6.8% phenylalanine, 6.5–6.7% valine, 4.6–7.5% proline, 4.6–5.9% arginine, 4.5% isoleucine, 3.9–4.0% threonine, 2.8–2.9% histidine, 1.8–1.9% cysteine, 1.5–1.6% methionine, and 0.7–0.8% tryptophan (John et al. 1979). The ideal diet should contain similar levels of amino acids to those found in larval tissues, with the exception of phenylalanine, which should be provided at 50% of the concentration in body mass, and of the limiting amino acids, threonine and tryptophan, which should be provided at twice the concentration found in larvae tissues.

Fats. Mealworm fat composition is rather constant when fed different diets rich in oleic, linoleic, and palmitic acids (Oonincx et al. 2015). While palmitic and oleic acids remain almost stable, independent of diet, possibly due to the synthesis of fatty acids ad novo by mealworms, linoleic acid may have to be supplied to diets (van Broekhoven et al. 2015). Increased ingestion of polyunsaturated C18 fatty acids lowers the proportion of C18 monounsaturated fatty acids in larval tissues (van Broekhoven et al. 2015).

The addition of lipid to dietary regimes is beneficial at low concentrations while high quantities are unfavorable and potentially pernicious (Morales-Ramos et al. 2013). While cholesterol is a necessary diet ingredient, a fat concentration above 1% does not benefit mealworm life cycle parameters (Fraenkel 1950) and becomes an inhibitory factor at concentrations >3% (Martin and Hare 1942). Moreover, high-fat foods promote the potential agglomeration of the substrate, resulting in lower aeration and movement of mealworms, thus negatively interfering with respiration (Alves et al. 2016). In addition, it has been proven that supplementation with 20%

lipid content increases the susceptibility to parasitism by nematodes (Shapiro-Ilan et al. 2008).

Carbohydrates. *Tenebrio molitor* growth is almost nil in the absence of carbohydrates. The optimal range is 80–85% (Fraenkel 1950). Although Fraenkel (1950) reported no significant differences in the growth of mealworms using glucose or starch, Davis (1974) observed less growth with starch, sucrose, or lactose than with glucose in diets containing amino acid mixtures. Similarly, bacteriological dextrin as a carbohydrate source induced weight gain of almost twice that achieved with glucose (Davis 1974).

The mealworm life cycle is highly influenced by the dietary protein-to-carbohydrate ratio (Martin and Hare 1942; Rho and Lee 2016; Urrejola et al. 2011). Rho and Lee (2016) reported an optimal protein-to-carbohydrate ratio of 1:1 for lifespan and lifetime reproductive success while Martin and Hare (1942) observed maximum growth at a minimum of 50% carbohydrate and a minimum of 15% (or more than 25%) protein in the diet.

The ingestion of diet consisting of several combinations of organic waste material (vegetables), yeast, and *Tenebrio* excreta results in body protein contents 2-fold higher and body fat content 5- to 6-fold higher than that of food, with a significant reduction in the values of crude fiber and carbohydrates (Ramos-Elorduy et al. 2002). Moreover, mealworms fed on low protein:carbohydrate ratios (0:42 and 7:35) diets have higher body lipid content (Rho and Lee 2014).

Vitamins. *Tenebrio molitor* shows no growth in the absence of vitamins of the B-complex (carnitine, thiamine, riboflavin, nicotinic acid, pyridoxine, or pantothenic acid) while there is growth, albeit slow, with the lack of biotin or pteroylglutamic acid (Hardouin and Mahoux 2003; Leclercq 1948; Martin and Hare 1942). On the contrary, the addition of vitamins A, C, D, E, and K to the diet provides no beneficial effect (Fraenkel 1950; Martin and Hare 1942). After mealworms reach half- or full-grown size, no addition of vitamins is required for the mealworms to complete larval development and to pupate (Leclercq 1948).

Nutritive Value and Use *Tenebrio molitor*

Tenebrio molitor larvae have been used for several feed and food purposes, from fish bait and feed for animals in captivity (Ghaly and Alkoaik 2009; Martin et al. 1976) to human consumption in many cultures (van Huis et al. 2013). For human consumption, mealworm larvae can be used fresh as a whole (Ghaly and Alkoaik 2009; Zhao et al. 2016) or as flour (Aguilar-Miranda et al. 2002). For animal feed, the flour can be used as a supplement in pig and poultry diet (Bovera and Piccolo 2015; Jin et al. 2016; Józefiak et al. 2016; Ramos-Elorduy et al. 2002; van Huis 2013; Veldkamp et al. 2012) and in aquaculture (Barroso et al. 2014; Belforti M. et al. 2014; Gasco et al. 2016; Henry et al. 2015; Ng et al. 2001).

Tenebrio molitor larvae, as in the majority of insects, are rich in protein with content between 43.3–66.8% dry mass (Ghaly and Alkoaik 2009; Jin et al. 2016). The amino acid profile shows the presence of almost all essential amino acids necessary for human consumption: 1.39–4.8% isoleucine, 2.81–8.65% leucine, 1.6–6.6% lysine, 0.64–7.6% methionine + cysteine, 3.99–13.05% phenylalanine + tyrosine, 0.93–4.43% threonine, 3.14–7.61% valine, 1.61–3.64% histidine, and 0–

1.8% tryptophan (Aguilar-Miranda et al. 2002; Barroso et al. 2014; Davis and Sosulski 1974; Ghaly and Alkoaik 2009; Jin et al. 2016; Jones et al. 1972; Ramos-Elorduy et al. 2002; Zielińska et al. 2015). Larvae also have a fat content between 17–42.48% dry mass (Adámková et al. 2016; Siemianowska et al. 2013). In relation to total fatty acids, there is a predominance of palmitic acid (9.33–23.7%), oleic acid (36.5–52.94%), and linoleic acid (3.8–33.58%) (Adámková et al. 2016; Aguilar-Miranda et al. 2002; Alves et al. 2016; Finke 2002; Jin et al. 2016; Jones et al. 1972; Martin et al. 1976; Zhao et al. 2016). While rich in protein and fat, the mealworm larvae are poor in calcium content, which can be problematic when exclusively feeding upon this insect (Klasing et al. 2000). The values of mealworm mineral composition range at 0.32–0.75 mg/g Ca, 1.45–3.4 mg/g Mg, 5.37–13.45 mg/g P, 6.7–13.8 mg/g K, 0.025–1.76 mg/g Na, 0.032–0.13 mg/g Fe, 0.012–0.04 mg/g Cu, and 0.082–0.145 mg/g Zn in a dry mass (Barker et al. 1998; Jones et al. 1972; Martin et al. 1976; Siemianowska et al. 2013; Simon et al. 2013; Zielińska et al. 2015).

Conclusions

Tenebrio molitor are a viable source of nutrition that may be used as a food or as a feed source or supplement. Its life cycle and relative ease of rearing make it amenable to mass production for such uses. Herein, we have reviewed available scientific literature to delineate the optimal environmental and dietary conditions for such mass production. The *T. molitor* studied lived its entire life cycle in the same diet and rearing system. We found that there are optimal temperature (25–28°C) and humidity levels (60–70% RH) for optimal growth and developmental time of *T. molitor*. Overpopulation in the rearing arena can inhibit pupation and incite cannibalistic behavior (Tschinkel and Willson 1971) and, thus, should be avoided. And, although mealworms can survive feeding exclusively on wheat bran, supplemental sources of protein (i.e., yeast, casein), fats (<1%), carbohydrates (80 to 85%), and B-complex vitamins improves growth rate and production of the colony.

Currently, there is little, if any, information on the microbiological (including zoonoses) or chemical risks resulting from the use and consumption of insects for food or feed. Environmental impacts are comparable to other animal production systems and can be decreased by the reuse of insect farming products and by-products (substrate and manure) (European Food Safety Authority 2015).

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