



# Sustainable and Cost-Efficient Feed Ingredients for Optimum Breeding of House Crickets (*Acheta domesticus*) for Human Consumption in Thailand

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Miljövänliga och kostnadseffektiva foder ingredienser för optimal uppfödning av hussysror (*Acheta domesticus*) för mänsklig konsumtion i Thailand

Lina von Hackewitz

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Swedish University of Agricultural Sciences, SLU

Department of Molecular Science

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Lina von Hackewitz

**Supervisor:** Sabine Sampels, Swedish University of Agricultural Science, Department of Molecular Science

**Assistant supervisor:** Rickard Engberg, Global Bugs AB Sweden

**Examiner:** Jana Pickova, Swedish University of Agricultural Science, Department of Molecular Science

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**Swedish University of Agricultural Sciences**

Faculty of Natural Resources and Agricultural Sciences (NJ)

Department of Molecular Science

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## Abstract

The world population is expected to increase over the next 30 years. The global food production system is under pressure to keep up with the growing demand. In the last years, increased attention has been paid to use livestock with lower environmental impact, like the house cricket *Acheta domesticus*. In Thailand's northern and north-eastern regions, entomophagy has been a tradition since the past. House crickets are farmed due to their capacity to utilise resources, more efficiently than traditional livestock. They can be fed with wide variety of plant material, such as byproducts and food waste from food industry and agriculture. The quality of the house crickets depends to a great degree on the quality, consistency and composition of the feed. To sustain optimal growth, cricket farmers are traditionally using high-protein chicken feed (approximately 21% protein). However, the chicken feed containing soy- and fish meal is not sustainable.

In this study some cost efficient and sustainable feed ingredients for soy- and fish meal for the in Thailand widely-used edible insect species, house cricket (*Acheta domesticus*), was investigated. The crickets were fed on feeds where soy- and fish meal was replaced with more sustainable products, such as food-industrial byproducts and weeds and which can be adapted to large-scale farm industry in Thailand. Three experimental feeds and one control feed were applied, to study the growth rate, survival rate and feed- and water use and feed conversion rate (FCR).

The results indicated that rearing the house cricket *Acheta domesticus* on byproduct is possible. However, it is important to fulfill the crickets need of nutrients to increase the growth and survival rate of the house cricket. Homogeneity and uniformity of feed is important to increase acceptability. Beyond the feed, farming temperature and relative humidity needs to be optimised, to increase the production of the house crickets.

**Keywords:** *Acheta domesticus*, house cricket, farming, feed, feed alternatives, chicken feed, growth, byproduct

## Sammanfattning

Världsbefolkningen förväntas öka under de kommande 30 åren. Det globala livsmedelsproduktionssystemet är under press för att hålla jämna steg med den växande efterfrågan på livsmedel. Under de senaste åren har det ägnats större uppmärksamhet åt att ersätta kött med lägre miljöpåverkan, som insekten hussysor *Acheta domesticus*. I Thailands norra och nordöstra regioner har entomologi varit en tradition sedan många år tillbaka. Hussysor föds upp för att de kan utnyttja resurser effektivt de kan matas med en mängd olika växtmaterial såsom biprodukter och matavfall från livsmedelsindustrin och jordbruket. Hussysors kvalitet beror till stor del på fodrets kvalitet, konsistens och sammansättning. För att upprätthålla optimal tillväxt använder cricketbönder traditionellt kycklingfoder med hög proteinhalt (ungefär 21% protein). Kycklingfodret är dock med soja- och fiskmjöl inte hållbart eftersom den har stor miljöpåverkan.

I den här studien studerades ett kostnadseffektivt och hållbart foderalternativ för soja- och fiskmjöl för i Thailand allmänt använda ätbara insektsarter, hussysor (*Acheta domesticus*). Sysorna matades med foder där soja- och fiskmjöl ersattes med mer hållbara produkter, såsom livsmedelsindustriella biprodukter och ogräs och som kan anpassas till den storskaliga jordbruksindustrin i Thailand. Tre experimentfoder och ett kontrollfoder applicerades för att studera tillväxthastighet, överlevnadshastighet och foder- och vattenanvändning och foderomvandlingsfrekvens (FCR).

Resultaten påvisade att uppfödning av hussysor *Acheta domesticus* på biprodukt är möjligt. Det är emellertid viktigt att uppfylla sysornas behov av näring för att öka tillväxten och överlevnaden. Fodrets homogenitet och enhetlighet är viktig för att öka acceptansen. Utöver fodret måste farmens temperatur och den relativa luftfuktigheten optimeras för att öka dess överlevnadstid.

**Nyckelord:** *Acheta domesticus*, hussysor, uppfödning, foder, foderalternativ, hönsfoder, tillväxt, biprodukter





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## Abbreviations

AdDV	Acheta domesticus densovirus
CrIV	Cricket iridovirus
FCR	Feed conversion rate
LCA	Life cycle assessment
RH	Relative humidity

# 1 Introduction

## 1.1 Global food challenges

The world population is expected to increase over the next 30 years from 7.7 billion people (2019) to 9.7 billion by the year 2050 (United Nations, 2019). The global food production system is under pressure to keep up with the growing demand for food (Tilman, 2014) which is estimated to grow with 70% during the next 30 years. Agricultural production leads to an estimated 80 percent worldwide deforestation (FAO, 2017). Loss of forest is a major source of greenhouse gases emissions and reduces plant and animal biodiversity. Reduction of biodiversity will reduce options for breeding new crops and plant varieties that may allow food systems to better adapt to climate change (FAO, 2017). Almost 60% of biomass harvested worldwide is for the purpose of feeding live stock (Cruz and Krausmann *et al.*, 2008). Increased global food production could lead to an increase of land degradation, greenhouse gas emissions, conversion of crops and cropland for non-food production, pest species infestation and overfishing of seas as well as an immense impact on air, water, and soil quality (Nellemann *et al.*, 2009). Feed production accounts for 60-80% of the greenhouse gas emissions for eggs, chicken and pork, up to the farm gate. For milk and beef that is 35-45% (Sonesson *et al.*, 2009). However, the acknowledgement of the environmental impact of livestock production has increased over the last years (Röös, 2019).



## 1.2 A prospective alternative

Edible insects are known for their competence to convert organic side streams (Oonincx *et al.*, 2015; van Huis *et al.*, 2017). This indicates that they can be fed with wide variety of plant material, such as byproducts and food waste from food industry and agriculture (van Huis *et al.*, 2017). In Thailand's northern and north-eastern regions, entomophagy has been a tradition since the past. Nowadays, the habit expanded nationwide, and insects are not perceived as food for poor people anymore. Almost 200 edible insect species are eaten in Thailand (Mongkolvai *et al.*, 2009) but only a few are produced and sold on regular markets (Nutrition Division, 1992). Increased attention has been paid to use livestock with lower environmental impact, like the house cricket *Acheta domesticus*.

House crickets are farmed due to their capacity to utilise resources, such as water and feed, more efficiently than traditional livestock (Oonincx *et al.*, 2015; Nagasaki *et al.*, 1991). With this ability, house crickets as alternative livestock production could lower the environmental impact (Halloran *et al.*, 2016; Oonincx *et al.*, 2012). They additionally have a greater growth efficiency and lower feed conversion rate (FCR 1.7-2.3) meaning less feed is needed to produce 1 kg of end product, when compared with traditional livestock (FCR 2.5-10) (Oonincx *et al.*, 2015; Tacon and Metian, 2008). With this knowledge, opportunities to use byproducts in farming of *Acheta domesticus* can increase the sustainability of the insects even more. However, the feed should not reduce the performance of the insects, including developmental rate, growth rate, survival rate and their nutritional quality for human consumption (Sorjonen *et al.*, 2019).

The quality of the house crickets depends to a great degree on the quality, consistency and composition of the feed. High quality feed has a direct impact on the economic viability of the farming process (Miech *et al.*, 2016; Van Huis *et al.*, 2013; Vantomme, 2015). Nutritional rich feed will convey to an increased dry mass of the crickets compared to a less nutritional rich feed (Chapman *et al.*, 2013). Therefore, it is essential to have a balanced diet for optimal growth of the house cricket because it will determine the growth rate, maturity rate, reproduction performance and the quality of the end product (Arganda *et al.*, 2014; Oonincx and van der Poel, 2011; Raubenheimer and Simpson, 2003).

### 1.3 Sustainable rearing

To sustain optimal growth, cricket farmers are traditionally using high-protein chicken feed (approximately 21% protein) (Durst and Hanboonsong, 2014; Gahukar, 2016). Equal to conventional livestock, the diet consists mostly of feed that could be consumed by humans (e.g. fish, beans, starch rich plants) which reduces the system efficiency drastically (Rosamond *et al.*, 2000). Soybeans and fish meal have been used in many years as a major protein source in feeds in the Thai cricket farm industry (Van Huisen, 2012) due to their high content, quality and digestibility of protein and amino acids (Cohen, 2004). However, both soybean and fish meal have a high environmental impact due to deforestation and overfishing respectively (DaSilva *et al.*, 2010). Unstable production and an increased demand of fishmeal led to increasing cost of aquaculture production (Tacon, 2005). An increase in soybean and oil prices has been seen due to rapid enlargement in demand caused by a growing world population, whereas growth in production has slowed down (Trostle, 2008). With world prices of feed ingredients increasing, the feed industry is looking for alternative protein sources.

In the cricket production, feed has a large impact on „Life cycle assessment, LCA , sustainability assessment (Smetana *et al.*, 2016). is a technique to Therefore, it is important to focus on maximising feed conversion and cricket growth rate as well as on improved sustainability of the feed (Berggren *et al.*, 2019). Using products in feed which are not possible to use as food for humans could improve the LCA assessment. This includes non-nutritive or unsafe food like agricultural and industrial byproducts, and feed crops to enhance local biodiversity (Lalander *et al.*, 2015; van der Fels-Klerx *et al.*, 2018).

### 1.4 Production site in Thailand, Global Bugs

Global Bugs Holding AB was founded in 2016 in Sweden. In the same year, Global Bugs Asia CO., Ltd was founded in Thailand for production of the house cricket *Acheta domesticus* as an alternative protein source for human consumption. In June 2018, Global Bugs Trading Co., Ltd. was established for processing, marketing and global sales for cricket products in Thailand. The ambition of Global Bugs is to produce high quality and low-cost crickets and products based on cricket powder. The facilities are situated in Hua Hin, Thailand, and have been certified and honored by the Board of Investment of Thailand. High standard leads to demanding global market standards especially for the European, North American and the Japanese markets.

Global Bugs has all the needed certification to approve the export of the house cricket (*Acheta domesticus*) from Thailand.

## 1.5 Aim of the project

The aim of this study was to find a cost efficient and sustainable feed alternative for soy- and fish meal for the in Thailand widely-used edible insect species, house cricket (*Acheta domesticus*). The crickets were fed on feeds where soy- and fish meal was replaced with more sustainable products, such as food-industrial byproducts and weeds, which can be suitable to large-scale farm industry in Thailand. In this study, three experimental feeds and one control feed were applied, to study the growth rate, survival rate and feed- and water use and feed conversion rate (FCR).

## 2 Background

### 2.1 The house cricket *Acheta domesticus*

In Thailand, rearing and selling crickets for human consumption and feed has been common for a long time (FAO, 2013). Previously, native cricket species like *Gryllus bimaculatus* DeGeer, *Teleogryllus testaceus* Walker and *T. occipitalis* Serville have been reared and marketed. Nowadays, farmers shifted to the house cricket *Acheta domesticus* (FAO, 2013), because it is easier to rear and has a more pleasant taste for the consumer than the native cricket species (Dust and Hanboonsong, 2014).

#### 2.1.1 Farming

There are three main farming strategies: wild harvesting (no farming), semi-domestication (outdoor farming) and farming (indoor farming) (Van Huis *et al.*, 2013). To retain the diversity of the crickets and provide a stable and sustainable food supply, semi-domestication and farming is the best way of farming. However, 92% of the globally produced insects, included house crickets, are wild harvested (Yen, 2015). Still, in recent years farming has increased (Jansson and Berggren, 2015).

As farming of house crickets became more popular, approximately 20 000 farms are running worldwide and most farms in Thailand are considered as medium – or large-scale enterprises (Gahuk, 2016; Hanboonsong *et al.*, 2013). A medium-scale farms produce around 120 to 3 000 kg crickets/year while a large-scale farm produces 4 800 to 12 000 kg crickets/year (Mott, 2017). On average, the estimated total world production of crickets is 7 500 tons per year (1996-2011) (FAO, 2013). Edible insects have long been seen as food for poor or rural people. Nowadays, even high-income earners are

consuming them. In Thailand, almost 200 edible insect species are reported to be consumed (Mongkolvai *et al*, 2009) but only few, including the house crickets, are predominantly consumed and sold regularly at markets (Van Huis *et al.*, 2013).

### 2.1.2 Breeding

In the past years, farming practices have changed and have been optimised to increase the yield in mealworm farming. There are four types of breeding containers which can be found in cricket farms; concrete cylinder pens, concrete block pens, plywood boxes and plastic boxes. The concrete cylinder pens are usually used for water drainage. One container can produce 2 to 4 kilograms of cricket and are approximately 80 centimeters in diameter and 50 centimeter high (Hanboonsong, 2013). They are commonly used in small- and medium sized farms but need a large space and cannot be moved easily. Second breeding container system are the concrete block pens, which can be found on many farms. The pen varies in size, depending on space availability, but the common size is 1.2 x 2.4 x 0.6 meters (Hanboonsong, 2013). Each pen can produce 25 to 30 kilograms of crickets, which is suitable for medium and large-scale farms. However, at a high-density cricket population, the risk of disease outbreak or overheating is high. Plywood boxes are the third container system in Thailand with a size of 1.2 x 2.4 x 0.5 meters (Hanboonsong, 2013). and in which 20 to 30 kilograms of cricket in each pen can be produced. They are easy to clean but the plywood boxes are sensitive to hot, cold or damp weather which can cause deterioration. The last container system is plastic boxes (Morales-Ramos, 2018). They can be stacked on a shelf and produce 6-8 kilograms of crickets which size of each plastic box can be 0.8 x 1.8 x 0.3 meters (Hanboonsong, 2013). In the past, the plastic boxes were mostly used for in small- and medium-sized farms but are nowadays also used for large-scale farms. However, the crickets in the top of the shelf have a higher death rate due to overheating (FAO, 2013). To increase the area and provide hiding space for the crickets, cardboard egg cartons are added to the breeding contains (Mott, 2017). At Global Bugs (2020), plastic containers of 100x50x50cm are used and 39 cardboard egg cartons, 30x30x5 cm big, with a weight of 0.63 kg each are added.

### 2.1.3 Life cycle

The house cricket's life cycle contains an incomplete metamorphosis also called 'hemimetabolous', which means that after the nymph develops from

the egg, the physiological body build is similar to the adult *Acheta domestica*. The life cycle includes egg, nymph and adult crickets. First, the in-top-soil deposited eggs are incubated in a separate room, where the temperature is 35 to 37 °C and the humidity about 80%. Given the right conditions, the eggs will hatch on day 8 (Global Bugs, 2020). Studies conducted on crickets show that the nymphs will hatch day 11 to 15 (Mott, 2017). After hatching, the small crickets, also called 'pin head', will be moved into another room, which has a temperature of 30-32 °C and a humidity of 60-70 % (Global Bugs, 2020). Booth and Kiddell (2007), however, show that the optimal temperature for the house cricket is around 30°C and the optimal humidity is 50-60%. The time of development is dependent on the temperature and humidity because the *Acheta domestica* is an ectotherm, which expresses that the metabolism does not produce enough heat to increase the body temperature. Therefore, the crickets are dependent on the surrounding environmental temperatures (Rantall *et al.*, 2002).

At Global Bugs (2020), there are three different hatching rounds. After 8 days, the first hatched eggs will be removed, the nymphs will be put into a big breeding box and will be placed into the production room. The eggs that did not hatch, are still in the incubation room. 20 hours after the first hatching, the second round of hatched nymphs will be removed. Thereafter, the eggs which did not hatch in the second round incubate another 20 hours in the incubation room before hatching. The third-round nymphs are put into a big plastic box and moved into the production room. This procedure is done to separate the fresh hatched nymphs and the non-hatched eggs and to avoid size differences when harvesting. If the nymphs are going to the production or mating room, is randomly chosen.

Different to other insects, *Acheta domestica* do not have a pupation phase. The crickets are only gaining in size and moult 7 to 10 times in their life (Clifford *et al.*, 1990). After the fourth or fifth moulting, differences between male and female house cricket can be seen. The female crickets develop a clearly visible ovipositor from the base of the females abdomen. The ovipositor is a dark, needle-like protrusion that will grow to two thirds the length of the body by adulthood, which is approximately day 36. There are also differences in size between the male and the female. The females are 15-25% larger than the males due to the eggs which the females are carrying (Mott, 2017).

Reaching the adulthood after 6-8 weeks (FAO, 2013), the male is starting to produce the characteristic mating chirp. The mating activity is starting within 24 to 72 hours after maturation, followed by females laying eggs

(Murthaugh and Denlinger, 1985). In this case, the ovipositor is used to lay the eggs 1 to 2 cm deep into the top soil. The laying activity is commonly 4 to 8 days after reaching adulthood (Murthaugh and Denlinger, 1985). The top soil container will be transported to the incubation room and the cycle will start over again.

#### 2.1.4 Harvest

At Global bugs, the crickets will be harvested at day 30. Previously, the harvesting day was at day 36, however, due to high mortality after day 30, the harvesting day was changed. Mating crickets are harvested after mating is finished, which is approximately day 40 (Global Bugs, 2020). In other farms, the harvesting day can differ between day 30 to 55 (CelAgrid, 2016).

When harvesting, egg cartons, feed and water will be removed, leaving only the crickets in the boxes. After 12 hours, the crickets are washed one time, to remove unwanted particles. The crickets are killed by freezing them at -18 °C for 25 hours (Global Bugs, 2020). In this process, the physiology of the house crickets is used. Due to that crickets are ectotherm; sub-zero condition leads to low-activity until the temperature rises again. This is also called *torpor*. If the sub-zero conditions last longer than 24 hours, then the crickets will not recover and die (Mott, 2017). After staying in the freezer for 25 hours, they are taken out and washed in different steps. In step 1, the crickets washes 7 minutes in filtered and UV sterilised water, in step 2, the crickets are washed for 2 minutes in filtered and UV sterilised water and in step 3, the crickets washed for 1 minute in filtered and UV sterilised water, which in total amounts to 10 min of washing. After the three washing steps, put into the microwave oven for 10 – 15 minutes at a temperature of 110-120°C to reduce the moisture content to approximately 5% and reduce microbial spoilage. In the final step, the crickets are milled or left whole for further processing (Global Bugs, 2019). In figure 1, the more detailed harvesting-process can be seen.

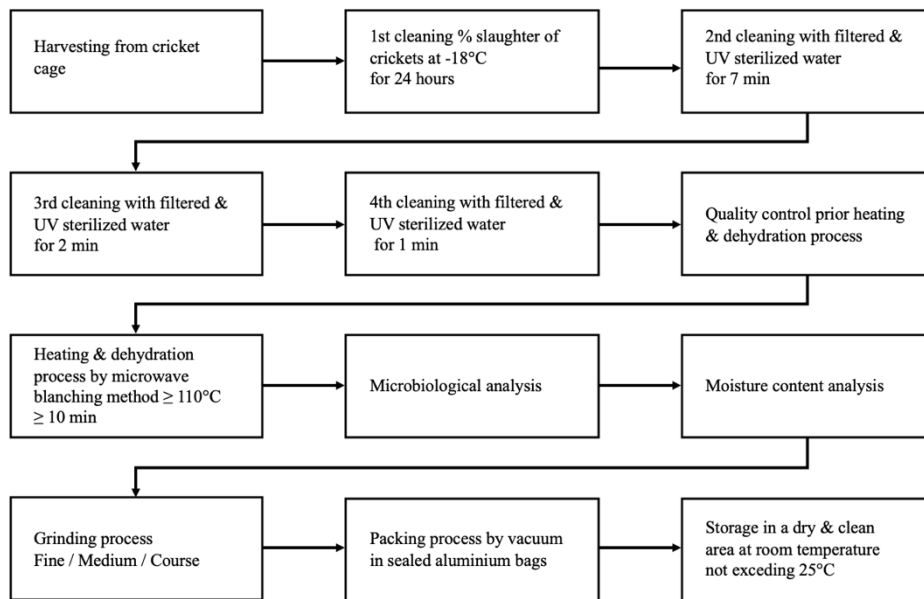


Figure 1. The harvesting process of the house cricket *Acheta domesticus* (Global Bugs, 2019).

### 2.1.5 Diseases

However, serious disease outbreaks have threatened the mass rearing of the house cricket. In commercially rearing facilities in North America and Europe, the *Acheta domesticus* densovirus (AdDV) caused problems (Szelei *et al.*, 2011; Weissman *et al.*, 2012). Additionally, cricket iridovirus (CrIV) caused problems in the cricket colonies in the past (Just and Essbauer, 2001; Kleespies *et al.*, 1999). Different viruses which can affect the mass rearing of the house crickets *Acheta domesticus* are shown in table 1. Beside viruses, crickets can be infected by other pathogens, for example fungi (Eilenberg *et al.*, 2015).



*Table 1. Insect-viruses which can infect Acheta domesticus.*

Virus	Susceptible stages	Pathobiology	References
<i>Acheta domesticus</i> densovirus (AdDV)	Last nymphal stage, young adults	Retarded grow, anorexia, lethargy followed by paralysation, finally death	Meynadier <i>et al.</i> , 1977 Styer and Hamm 1991 Szelei <i>et al.</i> , 2011 Weisman <i>et al.</i> , 2012
Cricket iridovirus (CrIV)	Nymphs and adults	Reduced fecundity and life span, apathy and disorientation, death	Kleespies <i>et al.</i> , 1999 Just and Essbauer, 2001 Jakob <i>et al.</i> , 2002 Adamo <i>et al.</i> , 2014
Cricket paralysis virus (CrPV)	Early- to mid-instar nymphs	Paralysis followed by death	Reinganum <i>et al.</i> , 1970, 1981 Morrissey and Edwards, 1977 Scotti <i>et al.</i> , 1981
<i>Acheta domesticus</i> volvovirus (AdVVV)	Unknown	Unknown	Pham <i>et al.</i> , 2013
<i>Acheta domesticus</i> mini ambidensovirus (AdMADV)	Unknown	Unknown	Pham <i>et al.</i> , 2013

## 2.2 Good Agricultural practices for crickets in Thailand

### 2.2.1 Farm design and layout

To prevent viruses and pathogens and produce house crickets of good quality and safety for the consumer, the Thai Agricultural Standard covers demand of good agricultural practices for cricket farmers. The standard covers farm components, feed, water, animal health, management of farm and environment (ACFS, 2017).

### 2.2.2 Farm management

To run a cricket farm, the location of the farm should be in a suitable area, with no risk of hazardous contamination affecting crickets and consumer safety. The house should be well ventilated and built with durable and easy to clean materials. The farm management manual should be available containing details on essential operation in the farm i.e. feed and water management, raising system, maintenance management and cleaning, environmental management, health management and record forms. For a good quality cricket management, the cricket breed should be selected, and clean shelters and materials must be used. It is also important to manage the cricket harvest in order to prevent contamination that will be harmful to consumers. Feed and water need to be clean to improve the health of the house crickets. It is also important that farms have staff who is properly trained and have knowledge about farming crickets. Furthermore, the staff needs to have a good personal hygiene and should receive health check-ups on a regular basis (ACFS, 2017).

### 2.2.3 Farm Animal health

To prevent diseases, disinfection and control of the equipment is required. Additionally, the entry and exit of visitors' should be traceable. To clean the farm, chemicals, disinfectants and hazardous substances registered at the Department of Livestock Development need to be used and instructions need to be followed. If a disease breaks out, or there is uncertainty of any incidence, Animal Epidemic Act and recommendations of Department of Livestock needs to be followed. Any disease treatment needs to be supervised by a veterinarian. To be aware of the environment, trash, garbage and cricket feces need to be managed with proper methods (ACFS, 2017).

## 2.3 Feed

Traditionally, cricket farmers use high-protein (approximately 21% protein) commercial chicken feed to raise their crickets. Nowadays, there has been feed formulated which are more specific for crickets (Durst and Hanboonsong, 2014; Gahukar, 2016). Fast growth, maturation and high survival rate are the important characteristics of production animals (Jensen *et al.*, 2017 (book)). To ensure offspring for the next generation, feed which promote high reproduction capacity is essential as well as large body masses to increase biomass production (Honek, 1993). It is therefore important to develop diets which meet species- and life stage-specific nutritional needs (Jensen *et al.*, 2017). Due to the need of high protein content in feed, the cost of the feed increases. Therefore, feed is one of the biggest issues when farming crickets (Van Huis, 2017). To reduce costs, some farmers are using the high-protein feed only until the crickets reach day 20. Thereafter, lower protein feed is used until harvest. Beyond protein, the house cricket needs other macronutrients, like carbohydrates including fibers and lipids. Additionally, micronutrients including vitamins, salts and minerals are essential in the diet. Oonick *et al.* (2015) and Lundy and Parrilla (2015) studies show that house crickets have equally or greater feed conversion than chicken or pigs when they are reared on well balanced organic side-streams of relatively high-quality feed.

### 2.3.1 Macro- and Micronutrients

To ensure that the house cricket has energy to grow and for configuration of chitin, the feed contains carbohydrates (Morales-Ramos *et al.*, 2014). Lipids, mainly polyunsaturated fatty acids such as linoleic and linolenic, are the main structural components of the cell membrane and additionally stored and supply metabolic energy during periods of sustained demand and help conserve water in arthropod cuticle (Payne *et al.*, 2016; Hogsette, 1992; Chapman, 1998). Amino acids, like leucine, isoleucine, valine, threonine, lysine, arginine, methionine, histidine, phenylalanine and tryptophan, are amino acids which cannot be synthesised. However, tyrosine, proline, serine, cysteine, glycine, aspartic acid and glutamic acid are amino acids which can be synthesised but synthesis in great quantities consumes a lot of energy (Costa-Neto and Dunkel, 2016; Lundgren, 2009). Essential micronutrients for the house crickets are vitamins, minerals and sterols, which cannot be synthesised (Cortes Ortiz *et al.*, 2016). For more detailed information about the requirements for edible insects, see table 2.

Table 2. Summary of the nutrient requirements for edible insects. Adapted from (Cortes Ortiz et al., 2016; Morales-Ramos et al., 2014).

Macronutrients			Micronutrients		
Minerals					
Carbohydrates	Lipids	Proteins	Stereols***	Vitamins	Elements** ***
Glucose *	Linoleic (Pfa) ***	Globulins	Cholesterol	A: Retinol + alpha and beta-carotene (Ls)	Hydrogen
Fructose *	Linolenic (Pfa) ***	Nucleoproteins	Phytosterols	B1: Thiamin (Ws)	Oxygen
Galactose *	Phospholipids ****	Lipoproteins	(beta-sitosterol, campesterol, stigmasterol)	B2: Riboflavin (Ws)	Carbon
Arabinose **		Insoluble proteins		B3: Nicotinamide (Ws)	Nitrogen
Ribose		Amino acids:		B4: Choline (Ws)	Calcium +
Xylose **		Leucin ***	Ergosterol	B5: Pantothenic acid (Ws)	Phosphorus +++++
Galactose **		Isoleucin ***		B6: Pyridoxine (Ws)	Chlorine
Maltose *		Valine ***		B12: Cobalamin (Ws)	Potassium +++
Sucrose *		Threonine ***		C: Ascorbic acid (Ls)	Sulphur
		Lysine ***		D: Cholecalciferol (Ls)	Sodium +++
		Arginine ***		Ergocalciferol (Ls)	Magnesium +++
		Methionine ***		E: alpha-tocopherol (Ls)	Iron ++
		Histidine ***		K: Phyloquinone (Ls)	Copper +++

Macronutrients		Micronutrients			
Minerals					
Carbohydrates	Lipids	Proteins	Stereols***	Vitamins	Elements** ***
		Phenylalanine ***			Zinc +++
		Tryptophan ***			Silicone
		Tyrosine ***			Iodine
		Proline ****			Cobalt
		Serine ****			Manganese +++
		Cysteine ****			Molybdenum
		Glycine ****			Fluorine
		Aspartic acid ****			Tin
		Glutamic acid ****			Chromim
					Selenium
					Vanadium

\*: Insects able to absorb and metabolize; \*\*: Insects able to absorb but not metabolize; Pfa: Polyunsaturated fatty acids; \*\*\*: Insects unable to synthesise, \*\*\*\*: Insects able to synthesise; Ws: Water-soluble; Ls: Lipid-soluble; \*\*\*\*\*: Listed in order of importance as essential for living matter (from top down). Minerals consist of combinations of cations and anions of elements; +++Important for insect growth; ++: Important in enzyme pathways including DNA synthesis, +: Important to a lesser extent, important role in muscular excitation.

### 2.3.2 Feed Conversion Ratio

Feed Conversion Ratio (FCR) is important as a decreased FCR will lead to increased use of feed per kilogram outcome. FCR varies depending on livestock and what kind of species it is. In table 3, the FCR of chicken, pork and beef compared to *Acheta domesticus* is given. There are few studies on edible insects. However, calculation of the edible portion (%) of house crickets might vary, due to that the calculations can be done on approximately 97% edible portion (whole cricket) or approximately 80% (without legs). Also, the exoskeleton is indigestible (approximately 3%) (Nakagaki & DeFoliart, 1991).

Table 3 shows that crickets are twice as effective in feed conversion as chicken, 4 times more efficient than pigs and 12 times more than cattle. According to several studies, the crickets convert feed more efficiently to body mass than conventional livestock. This is due to the fact that crickets are poikilothermic, which means that energy does not invest in maintaining body temperature constant (Van Huis, 2012), similar to fish and other aquatic organisms.

Table 3. Efficiencies of production of conventional meat and crickets

Country	House cricket <sup>a</sup> b	Poultry <sup>b</sup>	Pork <sup>b</sup>	Beef <sup>b</sup>
Feed conversion ratio (kilogram feed:kilogram liveweight)	1.7-2.3	2.5	5	10
Edible portion %	80	55	55	40
Feed (kilogram:kilogram edible weight)	2.1	4.5	9.1	25

<sup>a</sup>Oonicx *et al.*, 2015; <sup>b</sup>Smil, 2002

## 2.4 Feed alternatives

*Acheta domesticus* is a more sustainable protein source than livestock due to lower FCR and water intake (Oonicx *et al.*, 2015). However, the feed needs to be improved, to lower the impact on the lifecycle. Therefore, the soy and fish meal are replaced with different ingredients which are more sustainable, and which has a high protein and fat content. Below, the different ingredients replacements are explained and in table 4 the properties of each ingredient is listed.

Table 4. Properties of *Leucaena leucocephala* leaves, spent yeast, spent grain, biscuit by-product and tomato pomace.

Ingredient (DM 100%)	Crude Protein (g/100g)	Crude Fat (g/100g)	Crude Fibre (g/100g)	Ash (g/100g)
<i>Leucaena leucocephala</i> leaves	30	7.2	20	9

Ingredient (DM 100%)	Crude Protein (g/100g)	Crude Fat (g/100g)	Crude Fibre (g/100g)	Ash (g/100g)
Spent yeast	40	0.9	2.5	4
Spent grain	29	2.5	13	6
Biscuit by-product	11	8.9	1	2.7
Tomato pomace	25	16	60	2
Duckweed	27	3.5	20	20

#### 2.4.1 *Leucaena leucocephala*

*Leucaena leucocephala* (Lam) de Wit, is a leguminose browse plant which grows in tropical environment, including Thailand. It is known for its high protein (30 gram/100 gram) and fiber content (20 gram/100 gram) and is used as an ingredient in livestock feed. However, the leaves contain the amino acid mimosine, which is toxic to livestock when the level of 10% or higher is incorporated into the diet. High level will cause loss of appetite and therefore loss of weight and stunted growth, goiter and alopecia (NAS, 1977).

#### 2.4.2 Biscuit byproduct

Biscuit byproducts are the biscuits and biscuit rests which cannot be sold on the market due to different errors. The advantage of biscuits is the relative high fat content (12.3 g per 100 g) (Feed tables, 2020) which will be used as a replacement mainly for fish meal.

#### 2.4.3 Tomato pomace

Tomato Pomace is a byproduct which is produced in large amounts in the, for example, ketchup, juice and soup industry (Kaur *et al.*, 2008). It consists mainly of peels and seeds as well as a small amount of pulp (Lu *et al.*, 2019). Even if a large amount of tomato pomace is produced, it is often not properly utilised. However, sometimes it is used for pet and livestock feed and is known for its high fiber content (52 gram/100 gram) and relative high protein content (25 gram/100 gram). Additionally, the seeds enrich the tomato pomace with a high fat content (16 gram/100 gram).

#### 2.4.4 Spent yeast and grain

Ever since the urbanisation and civilisation of the Neolithic period, brewing and drinking beer has been a human activity (Meusdoeffer, 2009). To produce beer, large amounts of solid byproducts like spent grain and spent yeast are produced (Ajanaku *et al.*, 2011). Due to high protein, including essential amino acids, both spent grain and spent yeast are widely used as animal feed (Wang *et al.* 2001; Salama *et al.* 1995). Additionally, spent grain includes a high fiber content.

#### 2.4.5 Duckweed

Duckweed is known for its rapid growth and high nutritional value. The amount of protein is 27 gram/100 gram and crude fiber is 20 gram/ 100 gram. Duckweed has also a good amino acid quality where the content of leucine, theonine, valine, phenylalanine and lysine is high (Rusoff *et al.*, 1980). Also, the levels of the essential amino acids methionine and lysine is higher than found in other plant proteins (Gwaze & Mwale, 2015).

#### 2.4.6 Macro- and Micronutrient for feed alternatives

To improve the alternative feeds, it is important to have an as good amino acid and fatty acid composition as the fish- and soy meal. In table 5 the amount of different amino acids in fish- and soy meal compared with *Leucaena leucocephala* leaves, spent yeast, spent grain, biscuit by-product, tomato pomace and duckweed are shown. It can be seen that duckweed has a greater amount of amino acids



than fish- and soy meal and that spent yeast has a comparable high amount of each amino acid as soy meal.

Table 5. Amino Acids of fish meal, soy meal, *Leucaena leucocephala* leaves, spent yeast, spent grain, biscuit byproduct, tomato pomace and duckweed.

Ingredient (DM 100%)	Fish meal (62% protein) (g/kg) <sup>a</sup>	Soy meal (g/kg) <sup>a</sup>	<i>Leucaena leucocephala</i> leaves (g/kg) <sup>b</sup>	Spent yeast (g/kg) <sup>a</sup>	Spent grain (g/kg) <sup>a</sup>	Biscuit byproduct (g/kg) <sup>a</sup>	Tomato pomace (g/kg) <sup>a</sup>	Duckweed (g/kg) <sup>c</sup>
Lysine	50.7	24.2	33.9	29.2	9	4.2	10.5	56
Threonine	28.1	15.9	26.6	20.6	9	3.6	6.9	41
Methionine	18	5.6	9.8	7.1	3.9	1.8	3.9	16
Cystine	5.9	5.7	6.7	3	4.2	2.5	3.5	15
Methionine + Cystine	24	11.3	16.5	10.1	8.1	4.3	7.3	31
Tryptophan	6.6	4.9	n/a	5	2.9	1.4	2.1	32
Isoleucine	27.7	18.2	24.4	20.4	9.4	3.9	7.8	36
Valine	33.8	18.8	n/a	22.5	12.6	5.6	8.7	n/a
Leucine	48.5	29.4	44.4	28.3	18.1	8.1	13.3	78
Phenylalanine	26.5	19.7	28.3	16.5	11.6	5.3	10.3	47
Tyrosine	20.4	13.9	20.8	12.7	7.6	3.9	11.2	31
Phenylalanine + tyrosine	46.9	33.6	49.1	29.2	19.2	9.2	21.6	78
Histidine	17.6	10.7	12.3	10	5.6	3.4	6	18
Arginine	40.5	28.5	27.7	21.1	12	5.9	19.2	58
Alanine	43.1	16.6	31.1	30.8	12.3	4.8	8.8	69
Aspartic acid	61.7	43.3	86.4	37.7	17.7	6.5	20.5	120
Glutamic acid	84.6	68.5	64.0	68	47.7	25.1	29.1	114
Glycine	47.1	16.8	27.8	18.3	9.9	5.9	10.5	52
Serine	26.3	20.8	27.9	19.8	10.4	8.1	9.3	50
Proline	29.2	19.6	30.5	42.4	21.8	8.3	9.8	44

<sup>a</sup> feedtables.com (2020) <sup>b</sup>unpublished (1979) <sup>c</sup>Appenroth *et al.* (2018); n/a: no information available

As mentioned earlier, beyond protein and amino acids, the crickets need to get high quality fats in their diet. In table 6, the amounts of different fatty acids for fish- and soy meal compared with *Leucaena leucocephala* leaves,

spent yeast, spent grain, biscuit byproduct, tomato pomace and duckweed is shown.

*Table 6. Fatty acids of fish meal and soy meal compared with Leucaena leucocephala leaves, spent yeast, spent grain, biscuit byproduct, tomato pomace and duckweed.*

Ingredient (DM 100%)	Fatty acids (g/kg)	C16:0 (palmitic acid) (g/kg)	C18:0 (stearic acid) (g/kg)	C18:1 (oleic acid) (g/kg)	C18:2 (linoleic acid) (g/kg)	C18:3 (linolenic acid) (g/kg)
Fish meal <sup>a</sup>	7.7	13.6	2.8	9.4	1.6	1.5
Soy meal <sup>a</sup>	19.2	20.2	7.3	41.7	102	14.2
<i>Leucaena leucocephala leaves<sup>b</sup></i>						
Spent yeast <sup>a</sup>	3.2	-	-	-	-	-
Spent grain <sup>a</sup>	3.9	8.6	0.6	4.7	21.5	2.2
Biscuit by-product <sup>a</sup>		7.8	0.3	6.6	24.6	2.6
Tomato pomace <sup>a</sup>	6.6	9.3	4	15	35.6	1.3
Duckweed <sup>c</sup>	n/a	28.7	2.2	3.3	25	37.1

<sup>a</sup>feedtables.com (2020) <sup>c</sup>Appenroth *et al.* (2018); n/a: no information available

## 2.5 Cost efficiency

Feed is one of the highest investments in cricket farming, where it can account for as much as half of the total costs (Caparros Megido *et al.*, 2017). To lower the price, it is important to find a new source, which is as good as the high-protein commercial chicken feed (Dust and Hanboonsong, 2014). It is also important to lower the farming price to reduce the price per kilogram of the end product.

## 3 Materials and methods

### 3.1 Literature research

Information has been collected from scientific articles from databases as *Scopus*, *PubMed*, *Google scholar*. Additionally, information from the literature book *Insects as food and feed – from production to consumption* has been used. The papers investigated for this study are published between 1977 and 2020 and the used keywords are: *Acheta domesticus*, *house cricket*, *farming*, *feed*, *feed alternatives*, *chicken feed*, *growth*, *byproduct*.

### 3.2 Material

The study included 4 different diets of which 3 were experimental diets were soy- and fish meal were replaced. Crickets from the company Global Bugs in Hua Hin, Thailand were used in a pilot scale experiment at the farm (figure 2).



Figure 2. Ten days old crickets in the begin of the experiment.

### 3.2.1 Equipment

For this study, 12 plastic containers (32x22x19 cm) with aeration slits in the lid were used (figure 3). To increase the area and provide the crickets with hiding spaces, each container was furnished with 2 egg cartons. Each container included 15 grams of experimental feed on a round bowl (12 cm Ø) and 210 ml water in a plastic cup (220 ml) which were placed upside down on a paper tissue (12 cm Ø). Freshly hatched nymphs from the production line, where fed with cricket powder as ordinarily (Pride Pure, TFM Co., Ltd). At day 10, 100 cricket nymphs were randomised to each container and the experiment started. For each experimental feed, 3 replicates were used.



Figure 3. Plastic containers with lid used for the study. Different colors were used for different feeds: blue (feed b), Pink (feed a), green (feed c) and red (control feed).

### 3.2.2 Feed material

The replacements which were used are biscuit byproduct (Double Eagle; Brand Biscuits), *Leucaena leucocephala* leaves (Nippan Intercrop Co., Ltd.), tomato pomace (Mr. Udom), spent yeast (Tidjai Ltd., Part.), spent grain (Underdog Micro Brewery) and duckweed (Kalasinpanya Ltd., Part.).

The compositions of the control feed were given, and the ingredients of the experimental feed were calculated to reach the same composition. The compositions of feed a, b and c were followed: protein ( $22\% \pm 1.5$ ), fat (4%

$\pm 1$ ), fiber (5%  $\pm$  1.5) and ash (max 7 %). In table 7, the approximate composition of the experimental feed is shown.

*Table 7. The protein, fat, fiber and ash content of the control diet and 3 experimental diets (DM 87 %).*

Byproduct diet	Protein (%)	Fat (%)	Fiber (%)	Ash (%)	Proportion of replaced products in feed (%)
Control feed	22	4	5	-	0
Feed a	23.3	4.6	5.3	3.8	81
Feed b	22.4	4.0	5.8	4.1	78
Feed c	23.2	4.8	6.4	7.0	67

### 3.2.3 Diet preparation

To reach the same proximate composition of feed a, b and c, the amount of each ingredient were calculated and mixed together (table 8).

*Table 8. The composition of the 3 experimental diets (DM 87%).*

Diet	Corn meal (%)	Rice bran (%)	Biscuit byproduct <sup>a</sup> (%)	Leucaena leucicephala leaves <sup>b</sup> (%)	Tomato pomace <sup>c</sup> (%)	Spent yeast <sup>d</sup> (%)	Spent grain <sup>d</sup> (%)	Duck weed <sup>e</sup> (%)
Feed a	7	12	38	3	5	35	-	-
Feed b	13	9	34	3	-	-	41	-
Feed c	20	13	43	-	-	-	-	24

<sup>a</sup>Byproduct of biscuit production; <sup>b</sup>Byproduct of leucaena leucicephala production; <sup>c</sup>Byproduct of tomato production; <sup>d</sup>Byproduct of beer production; <sup>e</sup>Fast growing weed

Water was added to mix all ingredients properly. Thereafter, all ingredients were sundried and crushed with a mortar by hand until the feed reached powder state. The dry matter content of the experimental feed was 87% ( $\pm$ 3%). The control feed which were used is Pride Pure (TFM Co., Ltd). All

diets were stored in zipper bags at ambient temperature (30°C). Feed a, b, c and control feed can be seen in figure 4.



*Figure 4. Feed a, b, c and control feed on a plastic bowl (from left to right). Feed color: red-brown (feed a), beige (feed b), brown (feed c) and yellow (control feed).*

### 3.3 Experimental implementation

Feed and water were provided. At day 8 of the experiment, the crickets had increased in size which decreased the chance of drowning in a higher water density. Therefore, a plastic stick was placed under the plastic cup edge, to ensure increase of water flowing on the paper tissue and providing more water to the crickets (figure 5). All containers were checked every third day and feed and water were weighted and measured respectively. Feed and water were added as described below to ensure that the insects were al-

lowed to eat and drink *ad libitum*. The temperature and humidity were measured every morning (8am), midday (12am) and evening (17pm) and the photoperiod in this experiment was 12L:12D.

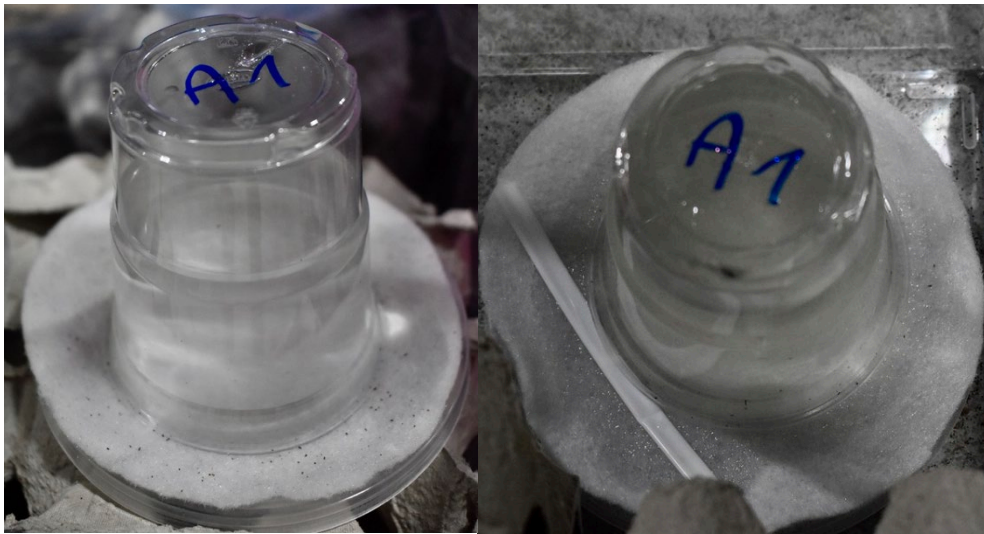


Figure 5. “Water spender” – a plastic cup upside down on a plastic bowl with paper tissue. Left without plastic stick to prevent small crickets from drowning and right with plastic stick to increase the water in paper tissue for bigger crickets.

### 3.4 Calculations

#### 3.4.1 Total weight

Each third day, 30 crickets from each container were weighed in groups of 10 individuals in a plastic cup (4 x 2.5cm). The weight of each individual cricket where calculated as:

$$\text{Weight of cricket} = \text{Total weight} - \text{Plastic cup weight} \quad (1.)$$

The mean weight of crickets in each container was calculated as:

$$\text{Weight of cricket} = \frac{\text{Total weight of weighted crickets from the container}}{\text{Number of crickets}} \quad (2.)$$

#### 3.4.2 Feed used

Every third day, the left-over feed was weighed, and feed was added to reach 15 grams of feed. Feed use was calculated as:

$$\text{Feed used} = \text{Feed provided} - \text{Feed left} \quad (3.)$$

#### 3.4.3 Feed conversion ration

Feed conversion ratio (FCR) is used to measure the feed conversion efficiency on dry matter (DM) basis and has been calculated as:

$$\text{FCR (\%)} = \frac{\text{Weight gained during feeding period}}{\text{Weight of feed consumed (DM)}} \quad (4.)$$



#### 3.4.4 Water used

Every third day, the water in the plastic cup was measured and refilled to 210 ml. The water use was calculated as:

$$\text{Water used} = \text{Water provided} - \text{Water left} \quad (5.)$$

#### 3.4.5 Weight gained

The 30-days old nymphs were weighted. The weight gain during the experiment was calculated as:

$$\text{Weight gained} = \text{Weight of 30 days old cricket} - \text{Weight of 10 days old cricket} \quad (6.)$$

#### 3.4.6 Temperature

The temperature was measured every morning and evening. The mean temperature was calculated as:

$$\text{Mean temperature} = \frac{\text{Temperature}}{\text{Number of measurements}} \quad (7.)$$

#### 3.4.7 Humidity

The humidity was measured every morning and evening. The mean humidity was calculated as:

$$\text{Mean humidity} = \frac{\text{Humidity}}{\text{Number of measurements}} \quad (8.)$$

#### 3.4.8 Survival rate

At the start of the experiment (10 days old nymphs) and at the end of the experiment (30 days old nymphs), the crickets were counted. The survival rate was calculated as:

$$\textit{Survival rate} = \text{Number of crickets (start)} - \text{Number of crickets (end)} \quad (9.)$$

#### 3.4.9 Cost efficiency

The costs of each experimental feed were calculated.

## 4 Results

### 4.1 Total weight

Already at day 8 of the experiment, the average weight of the crickets fed with control feed was higher than in feed a, b and c (figure 6). At day 30 (day of harvest), the average weight of the crickets fed with control feed was 325 mg, whereas the average weight of crickets fed with feed a was 101 mg, feed b 62 mg and feed c 59 mg.

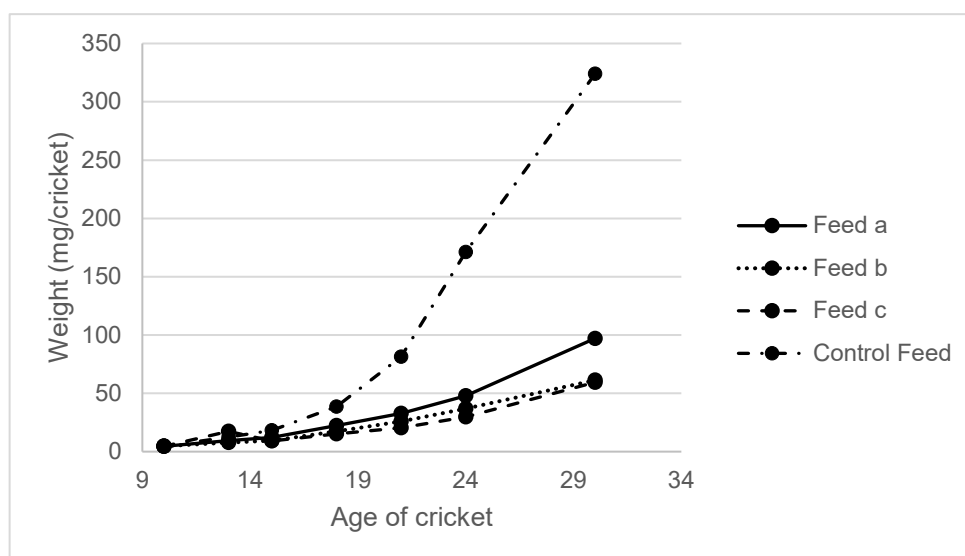


Figure 6. Weight of individual cricket fed with feed a, b, c and control feed from day 10 to day 30.

The greatest weight difference variation between crickets was found in the crickets fed with control feed where the weight range was 128 mg between crickets (min 264 mg; max 392 mg). The lowest weight difference between crickets was among crickets fed with feed b where the range was 65 mg (min 35 mg; max 100 mg). Crickets fed with feed a had a weight difference of 110 mg (min 38 mg; max 148 mg) and feed c 117 mg (min 33 mg; max 150 mg). In figure 7, the visible differences of randomly picked individual crickets fed with feed a, b, c and control feed is shown.



*Figure 7. Randomly picked crickets fed with feed a, b, c and control feed at age 30 days in plastic cup (from left to right).*

## 4.2 Feeding behavior and feed conversion rate

The feed conversion rate of feed a, b, c and the control feed can be seen in table 9. The greatest FCR has feed a followed by feed b and c. With 2.3, the control feed has the lowest FCR.

Table 9. *Efficiencies of production of Feed a, b, c and control feed.*

Ar	Feed a	Feed b	Feed c	Control feed
Feed conversion ratio (%)	38.1	28.6	26.6	2.3
Edible portion (%)	100	100	100	100

Differences in eating behavior between crickets fed with feed a, b, c and control feed can be seen in figure 8. Crickets fed with control feed tend to spread their feed in the whole box, whereas crickets fed with feed a and b this behavior was not observed. Crickets fed with feed b had a similar behavior as crickets fed with control feed, however, they only spread big pieces of spent grain in the box. The feed use is shown in figure 9.

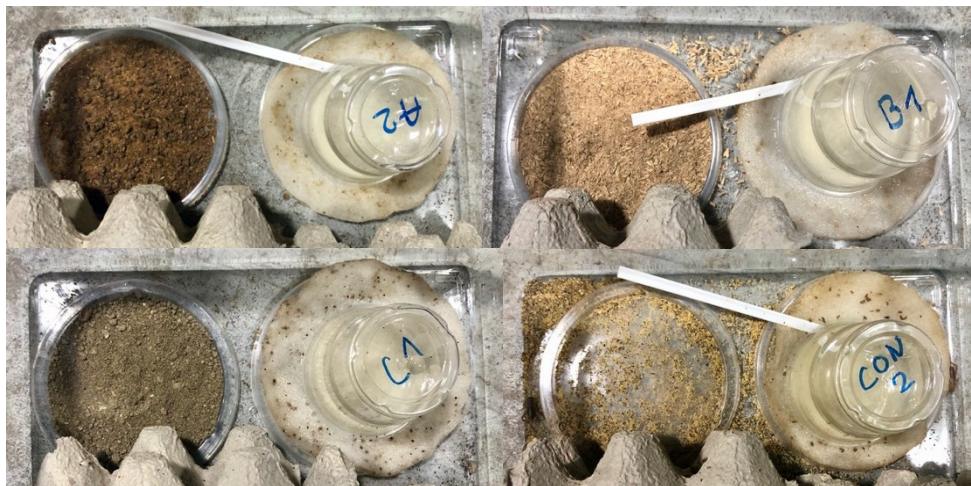


Figure 8. Feed a, b, c and control feed and “water-spender” after 3 days of use. Note: the plastic stick of feed c has been removed before taking the picture.

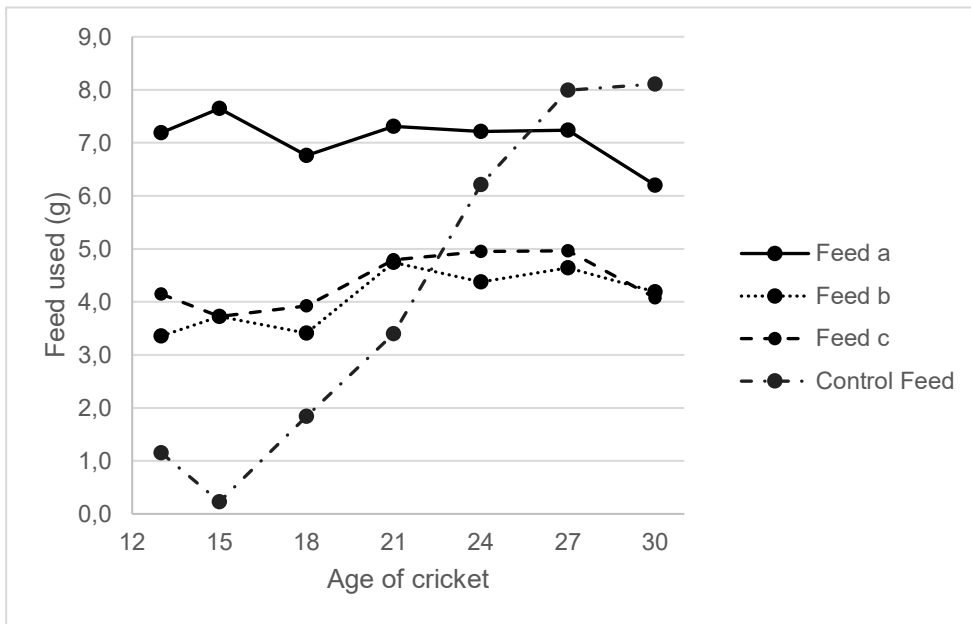


Figure 9. Feed used of crickets fed with feed a, b, c and control feed.

### 4.3 Water used

Water use of crickets fed with feed a, b, c and control feed is demonstrated in figure 10. With a water use of 295.2 ml, the control feed had the greatest water use in this study. Feed c is with a water use of 294.5 ml the greatest water use for the experimental feed followed by feed a (281.7 ml ) and feed b (277.0 ml).

In a smaller side experiment, the water evaporation has been tested. 100 ml water were added in the “water-spender” and left three day at the same place as the experimental boxes. Three days after, 89 ml of water was left, which means 11% of water evaporated. This result need to be considered in the water use.

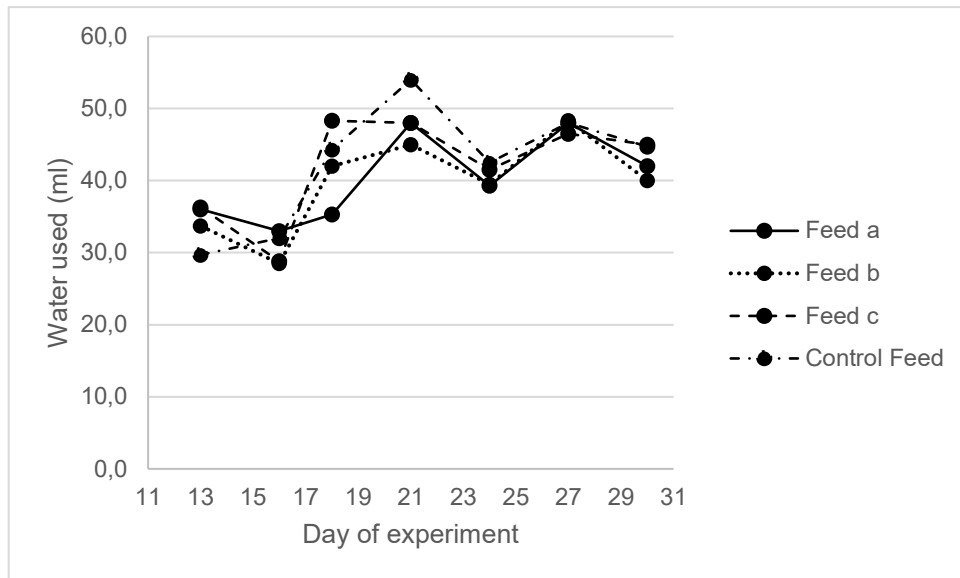


Figure 10. Water use of crickets fed with feed a, b c and control feed from day 10 to day 30.

#### 4.4 Temperature and Relative humidity

The temperature and relative humidity was measured three times a day: 8:00 am, 12:00 am and 5:00 pm. At 8:00 am, the average temperature was 32.4°C, 35.6°C at 12:00am and 35.1°C at 5pm. The total average temperature of this study was 34.4°C.

The relative humidity in this study at 8 am was 73.8%, 60.1% at 12 am and 60.8% at 5 pm. The total average relative humidity of this study was 64.9%.

#### 4.5 Survival

Mortality was high in crickets fed with feed a, b, c and control feed. However, the highest survival rate had crickets fed with control feed (40%) (figure 11). The lowest survival rate had crickets fed with feed a (18%) followed by feed

b (22%) and feed c (30%). Already at day 11 of the experiment, high mortality has been noticed at crickets fed with feed a and b.

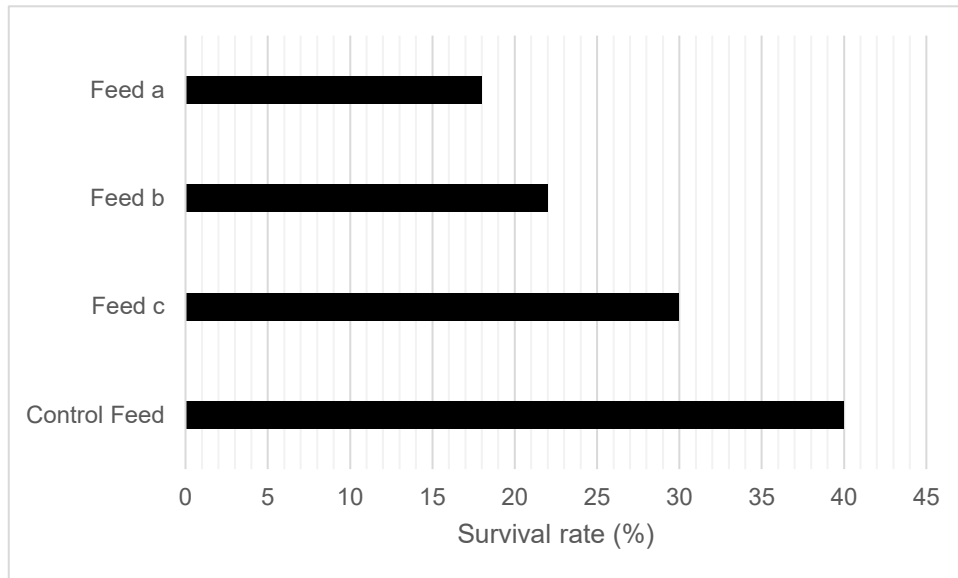


Figure 11. Mean survival rate (%) of the *Acheta domesticus* fed with feed a, b, c and control feed.

In the study, cannibalism had been observed between crickets which is shown in figure 12 and 13. In figure 12, a cricket eats another cricket who already was dead. In figure 13 (left), a large cricket attacks and eats a molting cricket and on the right picture (figure 13), a second cricket tries to steal the molding cricket from the large cricket. The large cricket on the other hand, runs away to escape with the molting cricket. Fighting behavior and cannibalism has been seen repeatedly on crickets fed with feed c, but also on feed a and b.





*Figure 12. Cannibalism between house crickets: the left cricket eats the right cricket.*



*Figure 13. Cannibalism between crickets: a large cricket eats another cricket which is molting (left). A smaller cricket tries to take the molting cricket from the large cricket (right).*

## 4.6 Cost efficiency

The total costs of feed a and b is 0.150 USD per kilogram (table 10). Feed c is with 0.25 USD per kilogram 1.7 times more expensive than feed a and b. The control feed is with 1.01 USD per kilogram 6.7 times expensive than feed a and b.

However, if calculating the total cost needed to produce one kilogram of crickets, the control feed is the most economical feed with 2.32 USD. Feed b is with 4.30 USD feed per one-kilogram end product the most economical feed of the experimental feed, followed by feed a (5.72 USD/1-kilogram crickets) and feed c (6.65 USD/1-kilogram crickets) (table 10).

Table 10. Total cost (USD) of Feed a, b and c compared with the control feed.

Diet	Corn meal (USD)	Rice bran (USD)	Biscuit by-product <sup>a</sup> (USD)	Leucaena leucicephala seed <sup>b</sup> (USD)	Tomato pomace <sup>c</sup> (USD)	Spend yeast <sup>d</sup> (USD)	Spend grain <sup>d</sup> (USD)	Duckweed <sup>e</sup> (USD)	Total costs/kg feed (USD)	Total costs/1kg crickets (USD)
Feed a	0.026	0.045	0.023	0.007	0.015	0.035	-	-	0.150	5.72
Feed b	0.049	0.034	0.020	0.007	-	-	0.041	-	0.150	4.30
Feed c	7.5	4.88	2.58	-	-	-	-	X	0.25	6.65
Control Feed	n/a	n/a	-	-	-	-	-	-	1.01	2.32

<sup>a</sup> By-product of biscuit production; <sup>b</sup> By-product of leucaena leucicephala production;

<sup>c</sup> By-product of tomato production; <sup>d</sup> By-product of beer production; <sup>e</sup> Fast growing weed; n/a: not available



## 5 Discussion

The main aim of this study was to determine the potential of using a more sustainable diet without soy- and fish meal for a large-scale industrial breeding of the house cricket *Acheta domestica* in Thailand.

### 5.1 Weight

Observing the average size and weight of the crickets, differences can be seen between control feed and feed a, b and feed c. An influencing factor of size can be the gender. Females are 15-25 % larger than the males (Mott, 2017), where many female crickets may increase the average of weight. Even if the weight range of the control feed was the highest, the differences of the cricket size fed with feed a, b and c were more visible with the eye (figure 6). Here, some crickets were as small as at the beginning of the experiment even if the feed was *ad libitum*. That could be due to the lack of proximate composition of nutritional content and the processing of the experimental feed. However, this is not confirmed.

### 5.2 Feed

#### 5.2.1 Macro- and Micronutrients

*Acheta domestica* is an omnivorous insect, which requires high protein and amino acid content in their diet (Chapman, 2013). However, Sorjonen *et al* (2019) study showed that feed made by plant-based byproducts and a protein content of 22% can be suitable for the *Acheta domestica*. Though, lower level of protein decreases the gain of individual biomass and slows down the development time in insects (Patton, 1967; Joern & Behmer, 1997). For growth and development of the crickets not only protein, but also carbohydrates and fats, are important (Chapman, 2013). The proximate composition of the experimental feeds were designed as similar as possible to the control feed. The proximate composition of nutritional content of the control feed was known by the macro nutrients; protein (21.5%), fat (4%) and fibers (5%) mentioned on the ingredients list. However, the percentage of each ingredient, which was included in the control feed, is a secret of the feed company. Therefore, the nutritional composition of the experimental diets has been developed by the using average values found in different literature studies. The in the study used ingredients might have a different than

the proximate composition of nutritional content what has been calculated. Furthermore, the control feed includes calcium carbonate, di-calcium phosphate, salts, minerals and vitamins, amino acids and preservatives which have not been added in the experimental feed. This may have an impact on the differences in results between the experimental feed and control feed, because extra nutrition was present in the control feed. The results show that the crickets fed with experimental feed had a lower weight and survival rate than crickets fed with the control feed.

In this study, feeds with a combination of a numerous ingredients has been tested trying to fulfill the needs of the crickets. More ingredients increase the complexity of each feed and makes it more challenging to evaluate the results. However, the calculation of the ingredients might be inaccurate, due to that the content has been taken from the literature.

In feed a, b and c, biscuit byproduct has been added to increase the yield of fat. The in the experiment used biscuits had a fat content of 8 grams per 100 grams. How much saturated and unsaturated fat is in the biscuits is unknown. Therefore, it is not possible to know why the weight of the cricket did not increase as fast as the control feed. Further investigations is required.

*Leucaena leucocephala* hay has been added to increase the protein content in feed a and b. The results shows that both feed a (18%) and b (22%) had a lower survival rate compared with feed c (30%). Even if the *Leucaena leucocephala* content in feed a and b is with 3% low, it might have affected on the survival of the crickets due to that contents greater than 5-8% can be toxic (NAS, 1977). Still, this needs to be further investigated.

In feed a, spent yeast was the main ingredient beside biscuit byproduct. It has a high amount of protein and has a comparable amino acid profile as soy meal (Feedtable, 2020). The purpose of tomato pomace in the feed was mainly to increase the protein and fat content and increase the amino acid profile content. The amino acid content can be as high as half of the amino acid of soymeal (Feedtable, 2020). Relative high quality and content of protein and amino acids in both spent yeast and tomato pomace could have increased the growth of the crickets because feed a resulted in the heaviest crickets compared with feed b and c. However, compared with the control feed the crickets are 3.2 times smaller than crickets fed with control feed.

Spent grain was the main ingredient in feed b and has a high amount of fibers (Wang *et al.*, 2001; Salama *et al.*, 1995) which could have supplied crickets with energy to grow and is used to configuration chitin (Morales-

Ramos *et al.*, 2014). It also includes important minerals (e.g. calcium, copper, iron and magnesium) and vitamins (Huige, 1994; Pomeranz and Dikeman, 1976). However, crickets fed with feed b and feed c had the lowest weight and only a survival rate of 22%.

In feed c, duckweed was included which can have a comparable amino acid profile as fish meal and a relative high amount of fatty acid. Also, in feed c, no *Leucaena leucocephala* had been added. This might explain the high survival rate of the crickets compared with crickets fed feed a and b. However, the crickets fed with feed c had the lightest weights in this experiment which could have been due to lack nutritional composition in this feed, because feed c had with 4 ingredients less ingredients than feed a (6) and feed b (5).

In this study, plant-based feeds only were fed to crickets. However, for crickets, both animal- and plant-based feed is an important source of nutrition (Tawes, 2014). Increased hunger or stress could lead to increased lust to fight (Nosil, 2002). Furthermore, if the feed does not fulfill the daily requirements, the crickets will continue to eat until their daily requirements are met (Hallett, 1995). Crickets with tendency to fight have been observed on crickets fed with feed c. The crickets started to attack each other when they were handled. However, the crickets have only been visually observed every third day. Therefore, it is unknown if this behavior would have been observable in between the measurement intervals as no observations have taken place. Nosil (2002) study shows that increased fighting success is given when the attacker is hungrier than the defender, which may lead to cannibalism. Crickets may only turn to cannibalism if there are hungry, which might refer to bad food supply. Bad food supply may additional explain the low survival rate in feed a, b and c. Behavior in crickets fed with feed a, b and c were observed, were crickets started to eat crickets which molted. In the moult-state, the crickets are more helpless because they are changing their skin. Due to paused body control, which is important for ability of competition (Marden and Waage, 1990; Plaistow and Siva-Jothy, 1996; Nosil and Reimchen, 2001), they might be easier to be attacked. To improve the feed and reduce the cannibalism thus mortality of crickets in the future, it is important to feed the crickets on adequate and high nutritious foods to implement survival and reproduction (Orinda, 2017).

### 5.2.2 Processing

In the experimental feed, homogeneity was not obtained as bigger particles were visible. Processing (e.g. mixing and particle size reduction) of the experimental feed which in the industry occurs with hammer or roller mill (Kiarie and Mills, 2019), has been implemented by hand. Quality of feed is determined by factors such as uniformity, homogeneity and size of feed particle distribution where uniformity and mixing homogeneity is one of the main challenges in feed manufacturing (Goodarzi Boroojeni *et al.*, 2016; Vukmirivic *et al.*, 2017). With this knowledge, the lack of uniformity, homogeneity and size of feed particle distribution in the experimental feed may have decreased the feed acceptance for the house cricket. Also, factors such as hardness and color can influence the quantity to consume and digest (El-Damanhour, 2011; LeBlanc, 2012). Therefore, the differences in color might have additionally an impact on the eating behavior (figure 8).

During the experiment, the experimental feed increased in weight even while the crickets consumed feed. The feed might have absorbed water due to high humidity of 64.9%. A side experiment showed, that in three days, the experimental feed gained weight with 23-48%. Therefore, an approximate feed use has been calculated. However, this data is still inaccurate. To improve future studies, the weight increase of both the feed fed to crickets, and the side study, need to be measured every day to have an increased accuracy of data. Also, the experimental feed has been mixed and dried without any industrial treatment and no additional preparations were used. In comparison, the control feed was received from the industry, which was industrially prepared and, in addition, included animal feed preservatives. This may explain the increased gaining in weight of the experimental feed in this study. However, further studies are needed.

### 5.3 Survival rate

The survival of the *Acheta domesticus* fed with byproducts was in this study 18-30%. These results are similar with previous studies who reported a survival rate of 6-80% in crickets fed with byproducts (Oonincx *et al.*, 2015; Collavo *et al.*, 2005; McFarlane 1962). However, the crickets in this study were only fed from day 10 to 30 of age. It needs to be considered that a lower survival rate could have been observed if the crickets were fed with the experimental feeds from day 1 of age. However, for crickets fed with control feed, the survival rate might be approximately the same (40%) because the diet did not change.

Crickets are difficult to handle when farming because they are moving fast (e.g. jumping). Behavior in farming and during the experiment shows that noise and vibration (e.g. knocking) leads to increased movement thus increased stress. During the measurements, some crickets jumped out of the box. This might lead to a lower number of crickets in boxes which can falsify the survival results. Furthermore, crickets might have jumped out of the box via the air holes. These aspects need to be considered in the results. To improve upcoming studies, the air holes need to be covered with a mosquito net or boxes with higher walls should be used to avoid crickets jumping out of the box. To reduce loss of crickets while rearing, harvesting or experimental measurements, crickets with reduced characteristics of jumping could be an option. This could be obtained through breeding or genetic manipulation.

Due to cannibalism of the crickets, no dead bodies were found. It is not proven how diet affects the frequency of cannibalism in cricket rearing situations. Therefore, it is necessary to further investigate this area in future studies.

#### 5.4 Feed conversion rate

The main variable of feed conversion efficiency for insects is determined by the feed composition (Scriber and Slansky, 1981). To fulfill the needs of crickets, a combination of different ingredients with suitable amounts of nutrients suitable for insects diet is needed.

Due to water absorption of the experimental feeds, increase of weight of the feed has been recorded. Even if the gained weight of the feed was measured and calculated, the feed used data is inaccurate. Especially for feed a was a weight gaining of 48% seen, which led to extreme feed use. However, a low growth and survival rate of the *Acheta domesticus* fed on the experimental feeds will result in higher FCR. With these extreme high FCR values, it cannot be compared with the result of the control feed or previous studies. However, similar results have been observed in the study of Vaga *et al.* (2019), where the final FCR values were much higher than in previous studies.

Oonix *et al.* (2015) study shows that the FCR of house crickets fed with a control diet had 2.3. Same result has been found in this study, where crickets fed with control feed had a FCR of 2.3. However, due to loss of feed in the box the FCR results for the control feed might be lower. Still, the result shows that house crickets had a comparable FCR as poultry and pigs (FCR



2.5 and 3.0) (Oonincx *et al.*, 2015; Tacon, 2008). Therefore, it is assumable that crickets can be reared with the same efficiency as common livestock.

## 5.5 Water use

With a water use of 295.2 ml, the control feed had the greatest water use in this study. This may be explained by the fact that the crickets fed with control feed were the heaviest crickets with an average of 324 mg per cricket. Furthermore, crickets fed with control feed had the greatest survival rate with 40%. High survival and heavy crickets may explain an increased water use. Feed c had with a water use of 294.5 ml the greatest water use for the experimental feed. The crickets fed with feed c had the lowest weight of all feeds, however, they had the highest survival rate with 30% compared with feed a and b. It needs to be considered that the evaporation of 11% is not included in the results.

On average, the crickets used 277 ml (feed b) to 295,2 ml (control feed) in this study. Some of the results had to be excised due to the egg cartons soaking up water which led to a lower water level in the water spender. The egg cartons were placed to not touch the water spender; however, crickets are moving which may cause vibration. The egg cartons tipped over and touched the water spender. To improve results for future studies, it needs to be considered that the egg carton cannot touch the water spender even if the crickets are moving.

## 5.6 Temperature and Relative humidity

For house crickets, there is no optimal temperature found yet. However, various studies have observed that a temperature range from 28-30 °C increase the growth of the crickets (Oonincx *et al.*, 2015; Sorjonen *et al.*, 2019). With a mean temperature of 34.4°C, the temperature in this experiment is 4.4°C higher than the optimum temperature. However, at 12 am, the average temperature was 35.6°C which is 5.6°C warmer than the optimal temperature. In nature, crickets can survive conditions well outside what is optimal. In boxes, controlling conditions are essential if the aim is to get a vibrant and strong colony (Mott, 2017). High temperature can increase the risk of stress (Mott, 2017) or over-heat, which have an impact on the survival of the crickets and may explain why the survival rate in this study was low.

With an optimal RH of approximately 50-60% (Booth and Kiddell, 2007), the average humidity in this study was 4.9% higher. Furthermore, the average RH at 8 am was 73.8% which is 10.8% higher than the maximum optimal RH. High temperatures and RH increases the risk of mold and mite invasion in the containers or facility, which may become a risk for both humans and crickets (Mott, 2017).

The temperature and RH in the farm depends on the outdoor temperature and the seasons. With minimal control over external factors in the farm, the temperature and the humidity can differ with many degrees respectively percentage. This, as mentioned, has a negative impact on the development and survival of the crickets. Temperature and RH are essential factors when farming crickets (Mott, 2017), where with optimal temperature and percentage, an optimal breeding might be reached. To optimise the temperature and RH and to decrease shifts between day, night and year seasons, an air-condition system can be installed to manage the issues. However, this might be a big investment which not all farms can afford.

## 5.7 Cost efficiency

Feed is a factor, which is one of the highest investments in cricket farming, which can be as much as half of total costs (Caparros Megido *et al.*, 2017). To lower the price for house crickets, it is important to lower the farming price including feed. With 1.01 USD per kilogram the control feed is less economical than the kilogram price for feed a and b (0.150 USD per kilogram) and feed c (0.25 USD per kilogram). However, FCR is an important measurement to see the actual feed needed to produce one kilogram of house cricket. Due to a low FCR of about 2.3, the control feed is with approximately 2.32 USD per kilogram cricket more economical than the experimental feed. Here, the price for feed a rose to as much as 5.72 USD per kilogram cricket. Feed b is with 4.30 USD per kilogram cricket the most economical feed of the experimental feeds, however, it is nearly double as expensive as the control feed. The most expensive experimental feed is feed c with 6.65 USD per kilogram of cricket. It is still important to be aware of that the FCR of the experimental feed is inaccurate, which leads to inaccurate prices. However, due to low growth and survival, it is logical that more of the feed needs to be used to obtain one kilogram of crickets.

In this study, small amounts of each ingredient were bought. Even if the kilogram price of the experimental feeds are low, a higher volume of each ingredient will further decrease the kilogram price of the experimental feeds.

Because, the higher the volume of each ingredient, the more the price will decrease (Karpoff, 1987). In cricket feed, many ingredients are same as in chicken feed. Chicken feed has been developed and optimised for a long time (Robinson *et al.*, 2003) and due to high production volume of each ingredient, the kilogram price is low.

To lower the price per kilogram of crickets, the weight and survival rate need to increase. Previous studies reported that the optimum protein level for cricket feed is 30% (Patton, 1967). However, a high protein content in the crickets diet increases the price. Therefore, a lower protein level will be used to lower the price of the feed. Previous studies show that even when the protein content is lower than 30%, the weight and survival rate can be improved (Sorjonen, 2019) which decreased the FCR and lowers the price per kilogram. Still, due to complications with the experimental feed and the weight gaining of it, the price might be lower than it is calculated. Low growth and low survival makes the feed still more expensive than the control feed.

## 6 Future studies and conclusion

In this study, the house crickets were fed on three different experimental feeds which contained different ingredients. Due to the complex nutritional needs of the house crickets, a combination of different ingredients increased the possibility to fulfill these needs. Therefore, protein, fat and fiber rich ingredients were used to assure the basic needs. However, many ingredients also increased the complexity to distinguish which ingredient or ingredients are inadequate to use in the feed. Due to that this study was a pilot study and lack of time, it was not possible to analyse the nutritional compositions in each feed. To draw further conclusions about the nutritional value of each feed, it would be recommended to analyse the nutritional value of the feed. This improves the knowledge of the nutritional feed compositions and may increase the possibility to make changes in the feed if it not fulfills the crickets' need.

The experimental feed gained in weight which did make it impossible to measure feed consumption. To improve future studies, dry feed can be changed every day and the percentage gained weight needs to be calculated of each day to get the actual feed consumption. Also, feed can be saturated with water before feeding to ensure that the feed is not gaining weight. Another option would be to improve the feed quality to make sure it do not gain in weight when having it in the farm at high humidity.

To improve the sustainability of cricket feed for large scale farming in Thailand and to be able to increase the weight and survival rate of crickets, the composition of the feed has an important role. Future studies might include animal sources which may increase the acceptance of crickets. Additional, studies on the behavior of cannibalism in relation to diet is of relevance to understand the relation to diet, cannibalism and survival rate

The conclusions are that rearing the house cricket *Acheta domesticus* is complex. In this study, the experimental feed based on byproducts and a fast-growing weed did most probably not fulfill the crickets need of nutrition. A lack of feed composition and other nutrients might be one of the attributes which may have influenced this result. Due to weight gaining of the feed the results has large drawbacks which also might have influenced the results. Also, absence of homogeneity and uniformity of feed a, b and c might have led to an decreased acceptability. Decreased acceptability can have led to decreased consumption. Beyond the feed, farming temperature and relative humidity is an important factor which needs to be consistent to optimise the rearing and outcome of the house crickets.



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## Appendix 1

### *A1. Nutritional value of Double Eagle Brand Biscuits.*

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Double Eagle Brand Biscuits (per 100 g)	
Calories	200 kcal
Sugar	6.67 g
Fat	8.00 g
Sodium	0.17 g

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## Appendix 2 – Popular Science Summary

### House crickets – How to make the sustainable more sustainable

*A rise in population, plus a rise in food demand, equals a higher carbon footprint. The humble house cricket may prove to be able to break this equation, especially when innovation never sleeps.*

In many countries, eating insects has been a tradition since the past. Edible insects are known for their high protein content and their delicious taste. The house cricket (*Acheta domesticus*) is one of the edible insects where the taste can be described as a whole roasted, nutty taste. They are farmed because their more efficient utilization of water and feed compared with traditional livestock. This means less feed and water is needed to produce 1 kilogram of end product. It is known that they can also be fed with byproducts and food waste from the food industry and from agriculture. Even though house crickets already are a better and more sustainable source of protein when compared with traditional livestock, it can be even better.



Equally to traditional livestock, the diet of the house cricket consists mostly of feed that could be

*Cricket-bread: Baked bread infused with cricket powder (serving suggestion) (Photo: Lina von Hackewitz).*

consumed by humans (e.g. fish, beans, starch rich plants). This reduces the system efficiency drastically. Soybeans and fish meal have been used for many years as

a major protein source in feeds of the cricket farm industry. It contains a high quality and digestibility of protein and amino acids. But both soybean and fish meal have a high environmental impact due to their contribution to deforestation and overfishing, respectively. An increased demand of fishmeal leads to increased overfished seas. An increase in soybean production leads to increased deforestation. Therefore, it is important to make house crickets, the already sustainable source of protein, even more sustainable.

In a recent study by Lina von Hackewitz, a master's student at the Swedish University of Agricultural Science in Uppsala, it was found that it is possible to use food industrial byproducts and fast growing weed as a replacement for soy- and fish meal. But the study showed also that the crickets fed with feed containing byproducts had a lower weight and

survival rate at the end of the trial. More feed was required to produce 1 kilogram of end product than crickets fed with feed containing soy- and fish meal. To prevent low weight and low survival rate, the composition of the feed is crucial to fulfil the daily nutritional need of the house cricket. Because of a low cost per kg of feed without the unsustainable ingredients, compared with the cricket feed, the amount of the experimental ingredients can be increased to boost the nutritional values of the experimental feed. This could better the growth rate and survival rate.

It is important to make the sustainable more sustainable, to always move forward with innovation. However, although the results were promising, more research is needed to improve the feed of the house cricket.