

Biodiversity and distribution of edible saturniidae (Lepidoptera) and their potential for mass rearing in Kenya

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Dedication

This work is dedicated to My sons, Leo and Luca, My husband, Kimani and My late mother Ms. Kusa.

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Abstract

Edible insects including saturniid caterpillars are traditionally consumed in sub-Saharan Africa. Saturniids are utilized both as food and a source of income for rural communities. This study includes a review of available information on edible saturniids in Africa. They feed on specific host plants and could either be univoltine or bivoltine depending on the location. For instance, *Cirina forda* (Westwood) feeds exclusively on *Vittelaria paradoxa* C.F. Gaertn (*Sapotaceae*) in West Africa while it prefers *Crossopteryx febrifuga* (Afzel.) Benth. (*Rubiaceae*) in Congo. They are attacked by parasitoids at different life stages. Local communities who consume them have vernacular names for different saturniid species signifying their cultural importance. Edible saturniid caterpillars are harvested in the wild which is unsustainable. Overharvesting and destruction of natural habitats point to the need to establish mass rearing protocols to ensure constant supply and reduce pressure on natural habitats. Communities process the harvested insects before consuming or selling within and outside their communities. Edible saturniids have an average protein content of > 50% and all the essential amino acids and an average fat content of >10%. Poor handling when processing can cause contamination of insects. Bacteria such as *Escherichia coli* and fungi such as *Aspergillus* spp. have been found in processed saturniid caterpillars hence the need for hygienic handling and proper post-harvest storage. Perceptions and practices among communities known to consume edible insects were additionally studied with particular emphasis on edible saturniids in Kenya. Popularity of the most and least preferred edible insects among the interviewees ranged from 88% for termites to 1.5% for lake flies respectively. Saturniid consumption is restricted to the coastal region among the Giriama community. Insect consumption varied by age, gender and occupation but was not influenced by the level of education nor region. Children and women were mainly involved in the collection and sale of edible insects. More than 98% of the respondents were familiar with saturniids, while only 67% were willing consumers. It is therefore necessary to create awareness on the nutritional benefits of edible saturniids and to process them into more palatable forms to alleviate malnutrition in the region. Seven edible saturniid species were identified including *Gonimbrasia zambesina* (Walker), *Go. krucki* (Hering), *Bunaea alcinoe* (Stoll), *Go. cocaulti* (Darge & Terral), *Go. belina* (Westwood), *Gynanisa nigra* (Klug) and *Cirina forda* (Westwood). They feed on 11 different host plants, and they all occur

twice a year except for *Go. cocaulti* which occurs only once. Two colour morphotypes were recorded for *Go. zambesina* and *B. alcinoe*. Predictive models revealed that tropical and subtropical regions were potentially suitable for *B. alcinoe* and *C. forda*. Climate change may affect their populations negatively by the year 2055. Parasitoids observed in this study belong to the orders *Diptera* and *Hymenoptera*. Larval pupal dipteran parasitoids included Tachinids like *Senometopia* sp. (cf. *evolans* Wiedemann) that emerged from *Go. zambesina*, *Go. krucki* and *Go. gueinzii*; a new *Ceromyia* sp. (Robineau-Desvoidy) from *B. alcinoe*, *Go. belina* and *Gy. maja*; a *Tachinidae* sp. from *C. forda* and a *Sarcophaga* sp. from *B. alcinoe*. The large Ichneumonid parasitoid, *Euryophion pisinnus* (Gauld & Mitchell) was also frequently encountered as a parasitoid of *B. alcinoe*. Braconid larval parasitoids constituted a *Cotesia* sp. from *Go. zambesina*, *Go. belina* and *B. alcinoe*; *Aleiodes trifasciatus* (Enderlein) from *Go. zambesina*; *Glyptapanteles maculitarsis* from *B. alcinoe* and a *Microgastrinae* sp. from *C. forda* were also recorded. Egg parasitoids included a parasitoid belonging to sub-family Entedoninae of Eulophidae and *Eupelmus* sp. (Eupelmidae) observed on *B. alcinoe* and *Go. krucki*, respectively. Thirteen bacteria species were isolated among which nine were potential entomopathogens. They include *Enterococcus mundtii* (Collins et al.), *Bacillus cereus* (Frankland & Frankland), *Staphylococcus sciuri* (Kloos et al.), *Staphylococcus gallinarum* (Devriese et al.), *Pseudomonas putida* (Trevisan), *Enterobacter hormaechei* (O'hara et al), *Enterococcus faecalis* (Andrewes & Horder), *Alcaligenes faecalis* (Castellani & Chalmers) and *Stenotrophomonas maltophilia* (Palleroni & Bradbury). Eight potential fungal entomopathogens were identified. Life cycles of *Go. zambesina* and *C. forda* were recorded. This study improves our understanding of edible Saturniids in East Africa and provides a baseline for the future development of mass rearing technics.

Zusammenfassung

Essbare Insekten, darunter Raupen von Saturniiden, werden traditionell in Subsahara-Afrika verzehrt. Saturniiden werden sowohl als Nahrung als auch als Einkommensquelle für ländliche Gemeinden genutzt. Diese Studie beinhaltet eine Analyse der verfügbaren Informationen über essbare Saturniiden in Afrika. Die Schmetterlingsraupen ernähren sich von bestimmten Wirtspflanzen und die Insekten können je nach Standort entweder univoltin oder bivoltin sein. *Cirina forda* (Westwood) beispielsweise ernährt sich in Westafrika ausschließlich von *Vittelaria paradoxa* (Gaertn C. F.), während sie *Crossopteryx febrifuga* (Afzel.) Benth im Kongo bevorzugt. Die unterschiedlichen Lebensstadien der Saturniiden werden von einer Vielzahl von Parasitoiden angegriffen. Lokale Gemeinschaften, benutzen einheimische Namen für die verschiedenen Saturniiden-Arten die sie konsumieren. Diese Namen reflektieren oftmals die kulturelle Bedeutung der Insekten für diese Gemeinschaften. Essbare Saturniiden-Raupen werden zumeist in der Natur wild geerntet, was eine nicht nachhaltige Praxis darstellt. Übernutzung und Zerstörung der natürlichen Lebensräume von Saturniiden unterstreicht die Notwendigkeit Verfahren zur Massenaufzucht der Insekten zu entwickeln, um so eine konstante Versorgung sicherzustellen und den Druck auf die natürlichen Lebensräume der Saturniiden zu reduzieren. Lokale Gemeinschaften verarbeiten die geernteten Insekten, bevor sie entweder direkt konsumiert oder später verkauft werden. Essbare Saturniiden haben einen durchschnittlichen Proteingehalt von >50 %, beinhalten alle essentiellen Aminosäuren und einen durchschnittlichen Fettgehalt von >10 %. Unsachgemäße Handhabung bei der Verarbeitung der Insekten kann zu Kontaminationen führen. Bakterien wie *E. coli* und Pilze wie *Aspergillus* spp. wurden in verarbeiteten Saturniiden-Raupen gefunden. Daher muss auf eine hygienische Handhabung und ordnungsgemäße Lagerung nach der Ernte großen Wert gelegt werden. Auch die Wahrnehmungen und Praktiken von Gemeinschaft die essbare Insekten konsumieren waren Gegenstand der Forschung dieser Untersuchungen, wobei essbare Saturniiden in Kenia hier im Vordergrund standen. Die Beliebtheit von essbaren Insekten unter den Befragten reichte von 88 % für Termiten bis zu 1,5 % für Seefliegen. Der Konsum von Saturniiden war auf die Giriama-Ethnie in der Küstenregion Kenias beschränkt ist. Der Insektenkonsum variierte nach Alter, Geschlecht und Beruf, wurde jedoch nicht

durch Bildungsgrad und Region beeinflusst. Kinder und Frauen waren mit 92,3 % bzw. 98,6 % hauptsächlich an der Sammlung und dem Verkauf von essbaren Insekten beteiligt. Mehr als 98 % der Befragten waren mit Saturniiden vertraut, jedoch nur 67 % erklärten sich als bereitwillige Konsumenten. Es ist daher notwendig, ein besseres Bewusstsein für die ernährungsphysiologischen Vorteile von essbaren Insekten zu schaffen und sie des weiteren in Konsumenten-freundlichere Produkte zu verarbeiten. Nur so kann ihre Akzeptanz erhöht werden, um so u.a. einen Beitrag zur Linderung der oftmals vorherrschenden Unter- und Mangelernährung in der Region zu leisten. Sieben essbare Saturniidenarten wurden identifiziert, darunter waren *Gonimbrasia zambesina* (Walker), *Go. krucki* (Hering), *Bunaea alcinoe* (Stoll), *Go. cocaulti* (Darge & Terral), *Go. belina* (Westwood), *Gyanisa nigra* (Klug) und *C. forda*. Ihre Juvenilstadien ernähren sich von 11 verschiedenen Wirtspflanzen und mit Ausnahme der univoltinen *Go. cocaulti* sind alle bivoltin. Für *Go. zambesina* und *B. alcinoe* wurden zwei Farbmorphotypen identifiziert. Vorhersagemodelle zeigten, dass tropische und subtropische Regionen potenziell für *B. alcinoe* und *C. forda* geeignet sind. Der prognostizierte Klimawandel könnte ihre Populationen bis zum Jahr 2055 jedoch negativ beeinflussen. Die in dieser Studie beobachteten Parasitoide gehören zu den Ordnungen Diptera und Hymenoptera. Zu den Diptera-Parasitoiden der Larvenpuppe gehörten die Tachiniden *Senometopia* sp. (cf. *evolans* Wiedemann), die sich aus *Go. zambesina*, *Go. krucki* und *Go. gueinzii* entwickelte; eine neue *Ceromyia* sp. (Robineau-Desvoidy) die auf *B. alcinoe*, *Go. belina* und *Gy. Maja* nachgewiesen wurde; sowie eine *Tachinidae* sp. aus *C. forda* und eine *Sarcophaga* sp. aus *B. alcinoe*. Eine große Ichneumoniden-Parasitoide, *Euryophion pisinnus* (Gauld & Mitchell), auch häufig in *B. alcinoe* beobachtet. Unter den Larvenparasitoiden aus der Braconiden Familie wurde eine *Cotesia* sp. auf *Go. zambesina*, *Go. belina* und *B. alcinoe* nachgewiesen; des weiteren wurden *Aleiodes trifasciatus* (Enderlein) auf *Go. zambesina*, *Glyptapanteles maculitarsis* auf *B. alcinoe* und eine *Microgastrinae* sp. auf *C. forda* beobachtet. Eiparasitoide beinhalteten eine *Entedoninae* sp. (Eulophidae) und eine *Eupelmus* sp. (Eupelmidae) die auf *B. alcinoe* bzw. *Go. krucki* nachgewiesen wurden. Insgesamt konnten 13 Bakterienarten isoliert werden, von neun potentielle Entomopathogene sind. Diese beinhalten *Enterococcus mundtii* (Collins et al.), *Bacillus cereus* (Frankland & Frankland), *Staphylococcus sciuri* (Kloos et al.), *Staphylococcus gallinarum* (Devriese et al.), *Pseudomonas putida* (Trevisan), *Enterobacter hormaechei* (O'hara et al), *Enterococcus faecalis* (Andrewes & Horder),

Alcaligenes faecalis (Castellani & Chalmers) sowie *Stenotrophomonas maltophilia* (Palleroni & Bradbury). Darüber hinaus wurden acht pilzliche Entomopathogene nachgewiesen. Des weiteren wurden die kompletten Entwicklungszyklen von *Go. zambesina* und *C. forda* erfasst. Diese Studie verbessert unser Verständnis von essbaren Saturniiden in Ostafrika und liefert die Grundlagen zur Entwicklung von Massenaufzuchtungsverfahren.

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List of acronyms and abbreviations

AUC – Area Under the Curve

CAR – Central African Republic

DNA - Deoxyribonucleic Acid

dNTPs - Deoxynucleoside triphosphate

DRC – Democratic Republic of Congo

FAO – Food and Agricultural Organization

GARP - Genetic Algorithm for Ruleset Production

GHG – Green House Gas

icipe - International Centre of Insect Physiology and Ecology

IUCN - International Union for Conservation of Nature

MaxEnt - Maximum Entropy

PCR – Polymerase Chain Reaction

PDA – Potato Dextrose Agar

SSA – Sub-Saharan Africa

UK – United Kingdom

UN – United Nations

USA – United States of America

WHO – World Health Organization

ZEF - Zentrum für Entwicklungsforschung

1.0 Introduction

1.1 The food security crisis

Food is a fundamental need for human existence, hence the second Sustainable Development Goal (SDG) ‘zero hunger’ target by the year 2030. Globally, about 820 million people suffer from hunger in Africa accounting for over 256 million people (FAO 2019). Apart from hunger, people also suffer from poor nutrition. Globally, over 150 million children under five are stunted (Development Initiatives 2018). Yet, at the same time, more than 600 million adults worldwide are clinically obese (Seidell and Halberstadt 2015). Food insecurity is mostly associated with micronutrients deficiencies and malnutrition. However, in some regions, micronutrient deficiency and access to calorie dense foods could lead to overnutrition (Development Initiatives 2018). This is often referred to as the “double burden” of malnutrition (DB). It is defined as the coexistence of overweight/obesity and undernutrition in the same country, community or household (WHO 2017). The prevalence of DB problem in households in Africa is below 5% (Wojcicki 2014). However, a higher prevalence has been reported in specific locations and populations in the continent. For instance, a prevalence of 43% has been reported among poor households in Nairobi, Kenya (Kimani-Murage et al., 2015).

Population trends suggest that the world population will increase from the current 7.2 billion to 9.7 billion by the year 2050 (UN 2019). The demand for food will rise by about 70% in the same period (FAO 2009). Yet, in a bid to feed this rapidly growing world population, environmental problems caused by food systems are increasing. Natural forests and wetlands continue to be cleared to pave way for pasture and crop production. This in turn leads to biodiversity loss and further accelerates man-made climate change (Clark and Tilman 2017; Williams et al. 2020).

While crop production has increased over the years, billions of people still lack nutritional security (Ramankutty et al. 2018). Diets consumed currently may contain the necessary calories but often lack important nutrients. The most common forms of nutritional insecurity is the lack of adequate protein or iron (Vos et al., 2017). Such inadequacies can have dire effects on child health and development. While small holder farmers produce almost half the calories consumed globally, they are more

likely to suffer nutritional insecurity. This implies that increasing crop yield alone may not be the solution to food insecurity.

One way of improving dietary diversity is by increasing meat production to ensure adequate animal protein supply. While meat is rich in protein and iron (Geissler and Singh 2011; Pighin et al. 2016), it has been associated with non-communicable diseases (Qian et al. 2020). More importantly, livestock production negatively impacts the environment accounting for more than 14% of total anthropogenic greenhouse gas (GHG) emissions (Gerber 2013). If the current trend continues, the livestock production sector may exceed the safe limits for human impact on the environment by the year 2050. With the current demographic growth rate, it is thus necessary to develop novel methods for or improve existing food production systems, especially with regard to animal proteins to ensure sustainability. Hence, one of the great challenges is how to produce more animal protein in a sustainable way.

Currently, the three main alternatives to livestock-based protein are microbial protein (Matassa et al. 2016), artificial/cultured meat (Stephens et al. 2018) and edible insects (Belluco et al. 2017; Patel et al. 2019; van Huis et al. 2015). However, making these alternatives truly viable requires extensive research on each stage of their economical and safe production and use.

1.2 Why edible insects

Edible insects have been considered as one of the possible solutions to food insecurity by providing an alternative protein source (Alemu et al. 2017; Gahukar 2011; Tanga et al. 2021; Tao and Li 2018; van Huis. 2013). Apart from proteins, edible insects are also rich in minerals, vitamins and antioxidants (Adepoju and Daboh 2013; Anvo et al. 2016; Ayieko et al. 2010; Di Mattia et al. 2019; Siulapwa et al. 2014; Tao and Li 2018). Recent studies have also shown that cricket consumption could be beneficial to the human gut (Stull et al. 2018). In addition to being a source of food, edible insects offer a source of livelihood for households in rural sub-Saharan Africa (Agbidye et al. 2009; Badanaro et al. 2014; Baiyegunhi and Oppong 2016; Moruakgomo 1996; Tanga et al. 2021).

Edible insects are a more sustainable protein source compared to the conventional sources like livestock. Their production utilises less land and water and produces less greenhouse gases (GHG) and ammonia compared to livestock production (Oonincx

et al. 2010; Oonincx and de Boer 2012). Insects convert feed better and have shown potential of being reared on biowaste which could in turn reduce production costs (Dobermann et al. 2019; van Huis 2013).

While the production of artificial meat and microbial protein is also deemed sustainable, edible insects have a distinct advantage; they have been consumed for over a millennium (van Huis 1996; van Huis et al. 2013). More than two billion people consume over 2000 species of edible insects globally (Jongema 2017). Entomophagy has been traditionally practised widely in Africa (Hongbété and Kindossi 2017; Mbata and Chidumayo 2003; Obopile and Seeletso 2013; van Huis 1996). Over 470 species are consumed in 54 African countries with 100 species being consumed in the East African region (Kelemu et al. 2015). The orders Lepidoptera, Orthoptera and Coleoptera dominate the list of edible insects in Africa. Edible caterpillars belonging to Lepidoptera lead with 60% of the edible insects in Africa (Kelemu et al. 2015; van Huis 2003). The main group of edible caterpillars consumed in Africa belong to the family Saturniidae (Latham 2015; Mabossy-Mobouna et al. 2016).

1.3 Edible Saturniidae

Saturniids, which constitute 27.5% of edible caterpillars (Mabossy-Mobouna et al. 2016) are the particular focus of this study. They are big colourful larval forms with spines on the surface. The larvae pupate into earthen or silken cocoons that form on the ground or on the tree stem from which brightly coloured moths emerge. The moths with a few exceptions are mostly characterized by large eye spots on the hind wings and a smaller glass spot on the front wings. Species are mainly differentiated by the general colour of the moth, the number and colour of rings surrounding the eyespots among other morphological traits (Pinhey 1956, 1972, 1975).

Edible saturniids are highly nutritious, rich in proteins, fibre and minerals (Braide et al. 2010; Dauda et al. 2014; Siulapwa et al. 2014). For instance, *Gonimbrasia belina* (Westwood) (Musundire et al. 2016), *Cirina forda* (Westwood) (Igbabul et al. 2015) and *Cirina butyrospermi* (Vuillet) have proteins constituting more than 50% of their proximate nutrient composition. Even better, most edible saturniids contain all essential amino acids (Paiko et al. 2014; Siulapwa et al. 2014; Yapo et al. 2017). Apart from nutritional benefits, edible saturniids provide income for many households in sub-Saharan Africa through their sale (Agbidye et al. 2009; Baiyegunhi and Oppong 2016;

Dube and Dube 2010; Gondo et al. 2010; Kozanayi and Frost 2002; Mbata and Chidumayo 2003; Ngute et al. 2020).

Edible saturniids occupy varied ecological zones in different parts of Africa. They can either be bivoltine or univoltine with the caterpillar occurrence coinciding with the rainy seasons (Ande and Fasoranti 1998; Latham 2015; Stack et al. 2003). They are also host specific, mostly feeding on forest trees, fruit trees and shrubs (Ngute et al. 2020). For example, *C. forda* mostly feeds on *Crossopteryx febrifuga* (Afzel.) Benth. (*Rubiaceae*) in Congo (Latham 2015) while it feeds on *Vitellaria paradoxa* C.F. Gaertn (*Sapotaceae*) in West Africa (Dwomoh et al. 2010; Odebiyi et al. 2011). *Gonimbrasia belina* which mostly occurs in the southern African region has the mopane tree, *Colophospermum mopane* (Kirk ex Benth.) Kirk ex J. Léonard (*Fabaceae*) as its primary host (Allotey et al. 1996; Dithlogo et al. 1996; Makhado et al. 2014). Their availability from the wild is associated with the prevalence of host plants and suitable seasons which is not consistent hence limiting their utilization as food.

Despite their numerous advantages, saturniids as a food resource faces a myriad of challenges. The main challenge is overharvesting and destruction of their natural habitats (Akpalu et al. 2009; Kozanayi and Frost 2002; Ngute et al. 2020; Thomas 2013). For instance, edible saturniid host plants are threatened by logging to produce timber. A good example is the African pearwood *Baillonella toxisperma* Pierre (*Sapotaceae*) which is a wild host plant of *C. forda* in Congo. The tree is used for timber and oil production from the fruits. The Africa pearwood is listed in the IUCN red list of threatened species, hence the need for its conservation (Ngute et al. 2020). While emphasizing on efforts to conserve natural habitats, it is also necessary to pursue avenues of mass rearing edible saturniids. This would reduce the pressure of wild harvesting and ensure a constant supply of the food resources instead of its current availability only during the suitable seasons.

1.4 Aim and Objectives

1. To establish community knowledge, practices, perceptions of edible insects in Kenya
2. To characterize the diversity of saturniids in Kenya using morphological and molecular methods
3. To determine the host range of selected saturniids

4. To map the abundance of saturniids both spatially and temporally across seasons in Kenya
5. To understand the constraints of selected saturniids in Kenya and assess the potential for mass rearing of selected saturniid species

1.5 Thesis structure

In chapter 2, I review the available literature on edible saturniid caterpillars in Africa. I use tailored searches with specific key words to understand the status of edible saturniid caterpillars. I have compiled a list of available edible saturniid caterpillar species and listed their vernacular names among different ethnicities. I have also described when and where they are available, documented their host plants and information on where they are consumed by whom. I highlight details on their harvesting, processing, cooking and trading, the role of gender in these activities and possible points of intervention to improve harvesting and processing methods. Food safety concerns related to edible saturniid caterpillars have been highlighted and possible methods to reduce contamination are discussed in this chapter. I have also discussed the cultural significance of edible caterpillars among communities including vernacular names and superstitions surrounding their presence and consumption is stressed. Lastly, I have discussed wild harvesting and the impact of overharvesting on saturniid populations and present the need to protect natural habitats of edible saturniids and possible interventions to conserve this food resource.

Chapter 3 sought to understand community perceptions and practices regarding edible insects in general narrowing down to edible saturniid caterpillars in Kenya. I went to communities with semi-structured questionnaires and sought to understand whether they consumed insects and if so, which ones. I further asked if they sell insects in local markets and if so, who collects and who sells in a bid to understand gender roles in insect collection and trading. To narrow down on saturniids, I asked whether they knew or had seen saturniids and whether they consumed them. I also went on further to find out if communities would be willing to rear saturniids given a chance, what would be their motivation and the challenges they would expect if they undertook such a project. Chapter 4 aimed at elucidating the ecology of edible saturniids in Kenya. It entailed identification of edible saturniid species and their host plants and understanding their distribution and seasonality. Morphological and molecular techniques were utilized.

Species distribution modelling was also carried out for select species to understand their distribution in Africa and the effect of climate change on future distribution.

Chapter 5 examines biotic constraints, especially parasitoids and microbial pathogens that affect saturniid populations in the wild. This chapter also presents my efforts at developing a mass rearing protocol for edible saturniids in captivity.

Chapter 6 is a discussion that synthesises all the findings of this study while chapter 7 concludes this study and highlights recommendations for future research on edible saturniids.

1.6 Study region

This study was carried out in Kenya. I chose to carry out my research in Kenya because although insect consumption has been traditionally practised in Kenya, previous studies in Kenya have focussed on other edible insects such as grasshoppers (Leonard et al. 2020), crickets (Magara et al. 2019), termites (Kinyuru et al. 2013) and lake flies (Ayieko et al. 2010). Information on edible saturniids caterpillars in East Africa is almost non-existent. Most of the research on edible caterpillars on the continent so far has concentrated on West, Central and Southern Africa (Agbidiye et al. 2009; Anankware et al. 2017; Latham 2015; Mbata and Chidumayo 2003; Ngute et al. 2020; Obopile and Seeletso 2013).

Different regions of the country were included to represent a mix of both insect consumers and non-consumers as well as different ethnic communities. In terms of agroecological zones, samples were collected from coastal Kenya to the highlands and to the semi-arid regions and the Lake Victoria basin. Siaya and Homa Bay represented the lake region, while Kilifi and Kwale represented the coastal region. Semi-arid regions included Machakos, Makueni, Taita, Kitui and Isiolo. The highlands included Murang'a, Embu, Meru, Nyeri and Nanyuki.

2.0 A review of Edible Saturniidae (Lepidoptera) caterpillars in Africa.

2.1 Abstract

Edible saturniids constitute an important component of traditional diets in sub-Saharan Africa. They are a source of livelihoods for many rural communities, both as food and as a source of income. This review compiles information on diversity, distribution, biotic constraints, nutrition and conservation concerns of edible saturniids. A compilation of nutritional profiles, amino-acids, mineral and fat content of saturniids is presented. Details of edible saturniids consumption and host plants are listed as well as ethnic names used for them in different parts of Africa. A comparison of collection, processing, storage and trading methods adopted by various communities is also included. Processing, which is mostly carried out by women, involves tedious and time-consuming methods that need to be improved. Poor handling and storage lead to bacterial and fungal contamination that raises food safety concerns. An in-depth discussion on conservation concerns and possible interventions is also provided. Further research on rearing edible saturniids is needed since wild harvesting is ecologically and environmentally unsustainable. Training women and youth on mass rearing technologies will ensure a more continuous supply of the insects and help preserve their natural habitats.

Key words: Saturniidae, edible saturniids, edible insects, entomophagy, edible caterpillars

2.2 Introduction

Food security is currently one of the major challenges in the developing world. The world population is expected to cross the 9 billion by the year 2050 resulting in a substantial increase in food demand (Grafton et al., 2015). There are about 820 million people suffering from hunger globally of which 256 million live in Africa (FAO, 2019). Apart from hunger, people also suffer from poor nutrition. Inadequate protein supply in developing countries is one of the causes of malnutrition (Ghosh et al., 2012). To meet the increased food demand, agricultural production will have to double by the year 2050 (FAO, 2009). Currently, agriculture occupies 30% of earth's land and increasing this proportion will conflict with other land uses. About 70% of the global agricultural land is utilized for livestock production especially for feed crop production

(Steinfeld et al., 2006). Increasing livestock production to nourish the growing population will require more agricultural land which is unsustainable as clearing more land will have huge detrimental effects on biodiversity and further accelerate climate change (Foley et al., 2005). Increasing livestock production also increases demand on water. Currently, 70% of fresh water is used for crop production, including fodder for the livestock sector (Doreau et al., 2012). In addition, CO₂ equivalents amounting to 18% of total anthropogenic greenhouse gases (GHG) and 64% of anthropogenic ammonia emissions are associated with livestock production (Steinfeld et al., 2006). Hence the need to explore alternative protein sources, such as edible insects that can be used as both human food and animal feed (Belluco et al., 2017; Chia et al., 2019; Dobermann et al., 2017; Henry et al., 2015; Kelemu et al., 2015; Makkar et al., 2014; van Huis, 2013). In addition to being highly nutritious (Charlotte et al., 2015; Di Mattia et al., 2019; Kwiri et al., 2014), edible insect production requires much less land and water and emits less ammonia and GHGs (Oonincx et al., 2010; Oonincx and de Boer, 2012) compared to other animal protein sources like fish, poultry and livestock.

Entomophagy (the consumption of insects) is traditionally practiced by 3,071 ethnic communities spread out in 130 countries across the globe (Ramos-Elorduy, 2009). About 2,100 species are consumed worldwide by over 2 billion people (Jongema, 2017). More than 500 species of edible insects have been recorded in Africa. Insects belonging to three orders namely Lepidoptera, Orthoptera, and Coleoptera are the most popular (Kelemu et al., 2015; van Huis, 2003). Lepidopteran insects belonging to 396 species from 36 families alone constitute 18.3% of the total edible insect species consumed by humans worldwide (Shockley and Dossey, 2014). In Africa, Lepidoptera constitute 60% of all edible insects consumed (Kelemu et al., 2015; van Huis, 2003). Among Lepidoptera, saturniid caterpillars with 109 species rank first and constitute 27.5% of edible caterpillars consumed. Edible saturniids mostly belong to the sub-family Saturniinae (Mabossy-Mobouna et al., 2016) but this review focuses on all edible Saturniidae in Africa.

2.3 Methods

A comprehensive compilation of literature on edible saturniids and their nutritional composition in Africa was established via searches on Web of Science, Medline and Google Scholar using the following search entry word combinations: (Genus and/or species name) AND ((edible OR edible insect OR entomophagy OR food OR feed)

AND (nutrition* OR protein* OR fat* OR mineral* OR Amino acid*)). To illustrate the distribution, conservation concerns and host plants of edible saturniids we searched for: (Genus and/or species name) AND (food plant OR host plant OR distribution OR habitat). For parasitoids and pathogens, search strings were: (Genus and/or species name) AND (parasitoid OR bacteria OR fungi OR entomopathogen OR food safety); while for collection, processing and trade, we used: (Genus and/or species name) AND (collection OR processing OR storage OR trade OR commercialization).

Means of nutritional components were calculated from values extracted from the available literature, tabulated, and utilized to generate graphs. All values for nutritional components were recorded on dry matter basis.

2.4 Diversity biology and distribution

Different species of edible saturniids are found in different parts of Africa. Although the seasons vary between regions, availability of the caterpillars depend on the weather (Ande and Fasoranti, 1997). They start to appear during the rainy season when trees have enough foliage for them to feed on. African saturniids have different host plants which mostly include forest trees, shrubs and some domesticated fruit trees (Agbidiye and Nongo, 2012; Anvo et al., 2016; Latham, 2015; Thomas, 2013).

For example, the Mopane worm *Gonimbrasia belina* Westwood is mostly found in southern Africa including Zambia, Zimbabwe, Malawi, Namibia, Angola, Botswana and South Africa (Allotey et al., 1996; Ditlhogo, 1996; Gondo et al., 2010; Kwiri et al., 2014; Thomas, 2013). The insect is widespread in areas where the main food plant, the mopane tree *Colophospermum mopane* (Kirk ex Benth.) Kirk ex J.Léonard (Fabaceae) occurs in forestland that cuts across southern Africa (Allotey et al., 1996). However, the mopane worm also feeds on other plant species (Appendix 1). *Gonimbrasia belina* is mostly bivoltine in Botswana, Zambia and South Africa with two generations per rainy season (Ditlhogo, 1996; Gondo et al., 2010). It usually occurs from October/November to December/January for the first and between February/March and April/May for the second generation (Ditlhogo, 1996, b). Adult moths live for about five days and their fecundity varies depending on location and time of the year (Ditlhogo, 1996). The moths are nocturnal, with males only flying near midnight, and hide or remain stationary during the day (Oberprieler, 1995). In the more arid areas of Namibia, *G. belina* is univoltine, occurring from February to April (Oberprieler, 1995;

Thomas, 2013) and in Zimbabwe between November and January (Dube and Dube, 2010).

Another popular saturniid is the shea tree caterpillar, *Cirina forda* Westwood. It can be found throughout sub-Saharan Africa (SSA) (Agbidye and Nongo, 2012; Badanaro et al., 2014; Dwomoh et al., 2010; Mabossy-Mobouna et al., 2016; Pinhey, 1956), and is the most economically important edible saturniid since it is consumed and traded in all the regions where it is found. In West Africa, *C. forda* feeds mainly on the shea tree *Vitellaria paradoxa* C.F.Gaertn. (Sapotaceae) (Ande and Fazoranti, 1997; Dwomoh et al., 2010; Odebiyi et al., 2011), while in the Democratic Republic of Congo (DRC) it prefers *Crossopteryx febrifuga* (Afzel. ex G.Don) Benth. (Rubiaceae) (Latham, 2015) though its host plant range is even wider (Appendix 1). For instance, it also feeds on the wild syringa, *Burkea africana* Hook. (Fabaceae) in Zimbabwe (Kozanayi and Frost, 2002) and both on the ordeal tree *Erythrophleum suaveolens* (Guill. & Perr.) Brenan (Fabaceae) and the African pearwood *Baillonella toxisperma* Pierre (Sapotaceae) in Cameroon (Ngute et al., 2020). *Cirina forda* is univoltine in Nigeria with the larval appearance coinciding with the rainy season between July and September (Ande and Fazoranti, 1997; Badanaro et al., 2014). However, it is bivoltine in DRC, where the larvae occur between November and January and October to May during the rainy season (Balinga et al., 2004; Latham, 2015). In Cameroon, *C. forda* and *Gonimbrasia epimethea* Drury occur in June-September, while *Bunaea alcinoe* Stoll, *G. alopia* Westwood, *G. obscura* Butler and *G. oyemensis* Rougeot in June to August (Ngute et al., 2020).

Another edible saturniid in the south Sudanian zone of Burkina Faso is *C. butyrospermi* Vuillot (Séré et al., 2018) that solely feeds on *V. paradoxa* (Appendix 1). Several saturniid caterpillars feed on the same host plant. For instance, in DRC, *C. forda*, *Lobobunaea phaedusa* Drury and *B. alcinoe* feed on *C. febrifuga*. The auri tree *Acacia auriculiformis* A.Cunn ex Benth. (Fabaceae) is also a food plant for *G. epimethea*, *B. alcinoe*, *G. oyemensis*, *G. eblis* Strecker, and *G. obscura*. Locals in DRC plant auri trees in their homesteads and raise saturniid larvae from the forest on them for later consumption. Trees that host multiple saturniids are especially useful for farming edible saturniids.

2.5 Factors affecting saturniid populations

Edible saturniid populations vary greatly year to year due to different factors (Latham, 2015). Especially natural enemies like predators, parasitoids and entomopathogens can seriously affect their populations. Parasitoids attack saturniids either at the egg, larval or pupal stage. More than 300 parasitoid species belonging to the families of Braconidae, Tachinidae, Pyralidae, Ichneumonidae, Sarcophagidae, Chalcidoidea and Proctotrupoidea (Peigler, 1994) have been recorded. For example, *C. forda* pupae are attacked by the chalcids *Hockeria crassa* (Walker), *Megaselia scalaris* (Loew) and *Hockeria* spp. (Dwomoh et al., 2004; Muhammad and Ande, 2014). *Hockeria* spp. have also been recorded as pupal parasitoids of *G. belina*. *Ceromya luteicornis* (Mesnil) attacks *G. epimethea* and *B. alcinoe* pupae. *Bunaea alcinoe* is also attacked by tachinid *Carcelia* spp., chalcid *Eucepsis* spp. and sarcophagid *Sarcophaga* spp. (Akanbi, 1973).

Most larval parasitoids of saturniids are braconids. For instance, *Glyptapanteles maculitarsis* (Cameron) has been recorded from larvae of *G. zambesina*, *B. alcinoe*, *G. maja* (Geertsema, 1975; Van Den Berg, 1971; Walker et al., 1990) and *G. epimethea* (Latham, 2015). The eupelmids *Mesocomys pulchriceps* (Cam.) *Eupelmus urozonus* (Dal.) and *Anastatus* spp., and the eulophid *Pediobius* sp. emerged from *G. belina* eggs in South Africa (Van Den Berg, 1971, 1974). In Nigeria *C. forda* eggs were parasitized by the eupelmid *Anastatus* spp. and the eulophid *Entedon* spp. (Muhammad and Ande, 2014; Odebiyi et al., 2011).

In addition to parasitoids, several fungal species have been isolated from *C. forda* in Nigeria, including *Aspergillus niger*, *A. flavus*, *Trichoderma* spp. *Fusarium solani*, and *Beauveria bassiana* (Muhammad and Ande, 2014; Odebiyi et al., 2011) as well as viruses like Granuloviruses (GV) and Nuclear polyhedrosis virus (NPV) (Muhammad and Ande, 2014). Finally, also birds and ants have been observed preying on edible saturniid caterpillars (Latham, 2015).

2.6 Attempts to mass rear edible saturniids

Since all the saturniid caterpillars consumed in Africa are harvested in the wild, it is necessary to delve into semi-domestication of the caterpillars which is widely practiced in Asia (Yen, 2015). Community farming of butterflies and saturniid caterpillars for income has been very successful in Tanzania (Morgan-Brown et al., 2010). Latham

(2015) gives an example of a farmer who brought live *C. forda* larvae from the market and introduced them in a savanna area that had plenty of *C. febrifuga* trees. He was able to harvest caterpillars in the subsequent seasons and reported that avoiding fires in the area kept the insects there every year. Locals in the Bas Congo province of DRC have planted *A. auriculiformis* trees near their homesteads where they place young caterpillars collected in the forest to mature for consumption. They also leave some of the caterpillars to mature and pupate in their compounds. The moths that emerge often lay eggs within vicinity of the home and the insect population grows (Latham, 2015). More work is required in creating awareness and training communities on such semi-wild production schemes of edible caterpillars to conserve the habitats and increase caterpillar yields.

2.7 Saturniids as food

Edible saturniid caterpillars are nutrient rich and hence form an important part of the diet for many people in SSA. The most popular species include *B. alcinoe*, *C. forda*, *C. butyrospermi*, *G. belina*, *G. epimethea* and *G. maja*. Most species are consumed in Congo and DRC (Appendix 1). While *G. belina* and *G. maja* are mainly eaten in southern Africa (Glew et al., 1999; Kozanayi and Frost, 2002; Kwiri et al., 2020; Mbata and Chidumayo, 2003; Obopile and Seeletso, 2013), *C. butyrospermi* is more popular in West Africa (Anvo et al., 2016; Ehounou et al., 2018; Séré et al., 2018), and *C. forda* is consumed throughout SSA (Badanaro et al., 2014; Latham, 2015; Mabossy-Mobouna et al., 2016; Ngute et al., 2020; Obopile and Seeletso, 2013).

In Nigeria, popular edible saturniids are larvae of *B. alcinoe* and *C. forda*, consumed mainly in Kwara, Benue and Niger states by the Igbo, Nupe and Yoruba people (Amadi et al., 2005; Braide et al., 2010; Fasoranti and Ajiboye, 1993). Despite a decline in entomophagy among the youth in Botswana, the mopane caterpillar remains highly sought-after compared to other edible insects (Nonaka, 1996; Obopile and Seeletso, 2013). A recent study in Côte d'Ivoire found *G. oyemensis* and *C. forda* as the most consumed and traded insect species in Abidjan (Ehounou et al., 2018), and *C. butyrospermi* is popular in the Sudanian zone of Burkina Faso (Séré et al., 2018).

In many communities the consumption of edible saturniids plays an important role in their culture. This is evidenced by local communities naming saturniid species in their ethnic languages and their ability to differentiate between species. Mabossy-Mobouna

et al. (2016) comprehensively documented the names of edible caterpillars in Congo, information that are lacking in many other African countries and which should be a key focus for future work on edible insects on the continent. For example, *B. alcinoe* is called 'Igu' by the Igbo people in eastern Nigeria and *C. forda* is called 'Kanni' or 'Munimuni' by the Yorubas in the west of the country (Amadi et al., 2005; Temitope et al., 2014). In southern Zimbabwe, the term mopane worm is used to refer to larvae of several saturniid species. They include *G. belina*, locally known as 'Macimbi', *G. maja* and *C. forda* ('Harati') named after their host plant, *B. africana* which is called 'Mukarati' in the Shona language (Kozanayi and Frost, 2002). In the Setswana language, widely spoken throughout southern Africa, *G. belina*, *B. alcinoe* and *C. forda* are referred to as 'Phane', 'Phata' and 'Nato', respectively (Obopile and Seeletso, 2013).

While some communities like the Teke community in Congo refer to several saturniid species with one name (here *B. alcinoe*, *G. alopia* and *G. eblis* are addressed as 'Inkele'), other communities in Congo such as the Baaka, Bomitaba, Kaka, Bodongo, Bonguili, Mbonjo and Yasswa all refer to *G. oyemensis* by the term 'Mboyoy' (Appendix 2).

2.8 Collection and processing edible saturniids

Harvesting, processing and storage of edible saturniids are similar across SSA. The tedious harvesting and processing are usually carried out by women and children. In Namibia, they constitute 85% of the harvesters. Occurrence of mopane worms coincide with the rainy season, hence the need for division of labour in the household with some members harvesting caterpillars while others till the land (Thomas, 2013). In contrast, men, women and children collect *C. forda* in equal measure in Togo, with the harvesting time coinciding with the school holidays, allowing the children to accompany their parents in the collection (Badanaro et al., 2014). As much as all community members keep an eye on the caterpillar season, children are the main insect collectors in the Bas Congo province of DRC. There, locals also carry the young caterpillars to their homesteads and place them on trees to mature (Latham, 2015). The San in the Kalahari Desert in Botswana set camp in the forest whenever the caterpillars are in season (Nonaka, 1996).

Caterpillars are collected by hand, either directly from the trees or from the ground while they climb down to pupate in the soil (Kozanayi and Frost, 2002; Mbata and

Chidumayo, 2003). In Nigeria, locals sometimes also dig the ground for prepupa that have burrowed to pupate. Moreover, they build pitfall traps around the base of the tree to trap the larvae prior to pupation (Agbidye et al., 2009; Agbidye and Nongo, 2012; Akanbi, 2002; Fasoranti and Ajiboye, 1993).

After harvesting, the gut contents are removed by holding the caterpillar between the thumb and index finger and squeezing the contents out through the rear end. This is done to every caterpillar individually, hence time consuming. However, when fully grown and ready for pupation, there is no need to squeeze the guts since bodies are filled with a highly nutritive yellow substance instead of plant material. In some cases, the caterpillars are starved for 1-2 days to empty the gut contents (Agbidye and Nongo, 2012; Akanbi, 2002; Kozanayi and Frost, 2002; Latham, 2015; Mbata and Chidumayo, 2003; Nonaka, 1996; Thomas, 2013). The squeezing process discolours and pricks the hands, and fingers of the handlers which can sometimes cause bleeding. In southern Zimbabwe, processors who can afford buy gloves while others cover their fingers with tree barks for protection. Some people use bottles to squeeze out the gut from several insects at once but the downside is that bottles exert too much pressure expelling the yellow substance preferred by consumers (Kozanayi and Frost, 2002; Thomas, 2013). Hence more effective, less laborious and less time-consuming processing methods that at the same time ensure the safety of the processors and retains the quality of the insects are needed.

In Nigeria, after gut cleaning, caterpillars are boiled and sun-dried and then they are ready for transport, storage and sale or direct consumption in households. For the latter, dried caterpillars are often fried in onions, pepper and salt, turning them into a delicious stew, often accompanied by cassava or other sources of carbohydrates (Agbidye et al., 2009; Akanbi, 2002; Fasoranti and Ajiboye, 1993; Temitope et al., 2014). In addition, caterpillars in Namibia are often also roasted on charcoal (Thomas, 2013). In Botswana (Nonaka, 1996; Obopile and Seeletso, 2013) and in Zambia (Mbata and Chidumayo, 2003), they are roasted in hot ash and sand before sun drying. In Zimbabwe, saturniid larvae are roasted on charcoal or cooked to facilitate removal of the spines; thereafter caterpillars are sun-dried. Alternatively, they can also be salted and sun-dried or boiled and then sun-dried. However, mopane worms processed in this way fetch lower prices in the markets since they still have the spines on. Moreover, salted worms have a whitish appearance not preferred by urban

consumers and they are usually sold at markets in rural regions (Kozanayi and Frost, 2002).

In the Bas Congo province of DRC, caterpillars are boiled in salted water with hot peppers, or in peanut butter, cassava leaves, or pumpkin and sesame seeds. When harvested in plenty, saturniid caterpillars are smoked and packed in sacks for later use or for sale in nearby markets (Latham, 2015). Although smoking allows for a longer shelf life (>3 months) compared to sun drying and boiling, the nutritional value of smoked and sun-dried caterpillars appears to be lower than fresh ones (Balinga et al., 2004; Rumpold and Schlüter, 2013).

2.9 Trading edible saturniids

Marketing of edible saturniid caterpillars is an important source of income for many households in SSA (Agbidye et al., 2009; Baiyegunhi and Oppong, 2016; Kozanayi and Frost, 2002; Moruakgomo, 1996; Yapo et al., 2017). However, owing to their seasonality, traders and vendors do not rely solely on these insects for income but are also involved in farming and trading of other food commodities (Balinga et al., 2004; Ngute et al., 2020).

Some traders buy caterpillars in bulk for sale in urban cities or for cross-border trade (Obopile and Seeletso, 2013; Thomas, 2013). For instance, in Zimbabwe, mopane worms are packed in sacks or large tins and sold to traders who repackage them into smaller quantities for resale on local markets. There they are either repackaged in plastic bags or wrapped in old newspapers. In some cases, bulk traders blend high quality and low-quality worms to fetch a better price (Kozanayi and Frost, 2002). The most traded saturniid in this region is clearly *G. belina*, though other species like *G. maja*, *C. forda* and *G. ertli* are also available but they are comparatively less popular. Again, traders often mix the less popular, and by consequence also less expensive, species with *G. belina* caterpillars since it is difficult to market them individually (Kozanayi and Frost, 2002).

Several studies looked into the marketability of edible caterpillars in different parts of Africa. However, most value chains of edible saturniid caterpillars are still in their infancy. For instance, in Côte d'Ivoire, *C. forda* and *G. oyemensis* are collected by insect collectors in forests (both national and those in neighbouring countries), then sold to local traders, followed by wholesalers, and eventually are marketed by retailers to rural and urban consumers, which can be lucrative. In 2018, a kilo of *C.*

butyrospermi dried caterpillar was valued at 3.5 USD or a box of 750 g of *G. oyemensis* dried larvae at 6 USD on urban markets in Abidjan, and with average monthly income from sale of insects >100 USD and profit margins of up to 69% for traders (Ehounou et al., 2018).

In Togo, while collection involves men, women and children, *C. forda* trade is almost entirely carried out by women. The trade chain involves, wholesalers, middlemen, and retailers, and because of their seasonality women sell the caterpillars alongside other commodities like grains and legumes (Badanaro et al., 2014).

In Zimbabwe, mopane worms are sold in both rural and urban areas, with women being involved in small scale retailing and vending. However, bulky cross-border transportation and wholesale is mainly dominated by men. The caterpillars can be found in markets, supermarkets, roadside stalls, bus terminals as well as hawked in beer halls as a snack. In supermarkets, they are well packed and labelled. Cross-border trade occurs mainly in South Africa, Zambia, Botswana and even in DRC (Kozanayi and Frost, 2002).

The most traded saturniid caterpillars in Namibia are *G. belina* and *G. maja* which are generally referred to as mopane worms. They play a crucial role in poverty alleviation for poor families, through trade, and providing food security, via direct consumption, for poor families. While harvesting is mostly carried out by women and children, unemployed males are often involved in the trade on urban markets. In Zimbabwe and Namibia, cross border trading of mopane worms to countries such as Angola is important (Thomas, 2013).

2.10 Nutritional composition of edible saturniids

A literature comparison on the nutritional content of five saturniid species (*C. forda*, *B. alcinoe*, *G. belina*, *G. maja* and *C. butyrospermi*) showed that proteins account for the largest percentage of the proximate composition of edible saturniids. All five species have an average protein content > 50% (Figure 1). The average fat content is >10% with a range of 11.5%-17.92% (Appendix 3). *Bunaea alcinoe* caterpillars have the highest fat content, and the ash content is ranging between 5% to 7.3% (Appendix 3). *Cirina forda* has the highest mean carbohydrate content (12.63%), while *B. alcinoe* has the lowest (3.16%), though the latter value is based on only one study (Agbidye et al., 2009). Mean fibre content is highest for *G. maja* and *G. belina* and lowest in *B. alcinoe* (Figure 1). All nutritional content studies for *C. forda* were carried out in West

Africa with the saturniid feeding on shea trees (Agbidye et al., 2009; Badanaro et al., 2014; Igbabul et al., 2015; Omotoso, 2006; Paiko et al., 2014). More studies are required to assess the effect of other host plants on the nutritional composition of *C. forda* larvae.

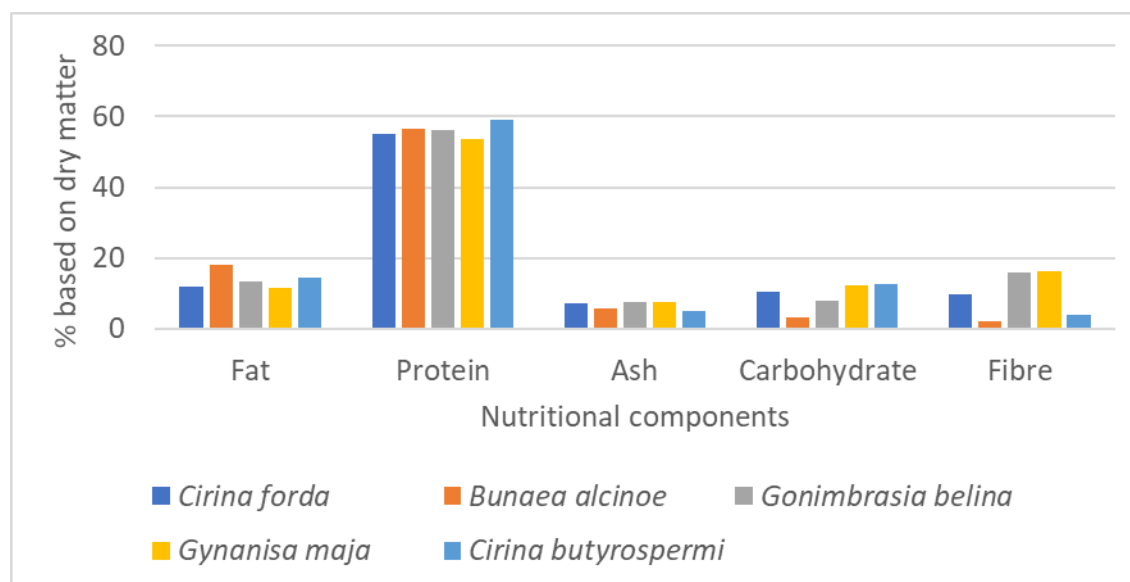


Figure 1. Average nutrient content (based on dry matter) of edible saturniids from literature

Saturniids also contain all the essential amino acids (Figure 2). The reported means of histidine, phenylalanine, tyrosine and threonine exceed the recommended daily allowance for adults (WHO, 2007) for the above five edible saturniid species (Figure 2). For *B. alcinoe*, amino acid contents exceed requirements for adults for all essential amino acids, while *C. forda* exceeded all except for histidine. No data could be retrieved on phenylalanine and tyrosine levels. Although amino acid levels for *G. maja* and *G. belina* larvae fall below the daily requirement for adults for isoleucine, leucine, lysine, methionine cysteine and valine, they still provide a substantial amount of amino acids (Figure 2). Edible saturniids are also rich in non-essential amino acids (Appendix 4).

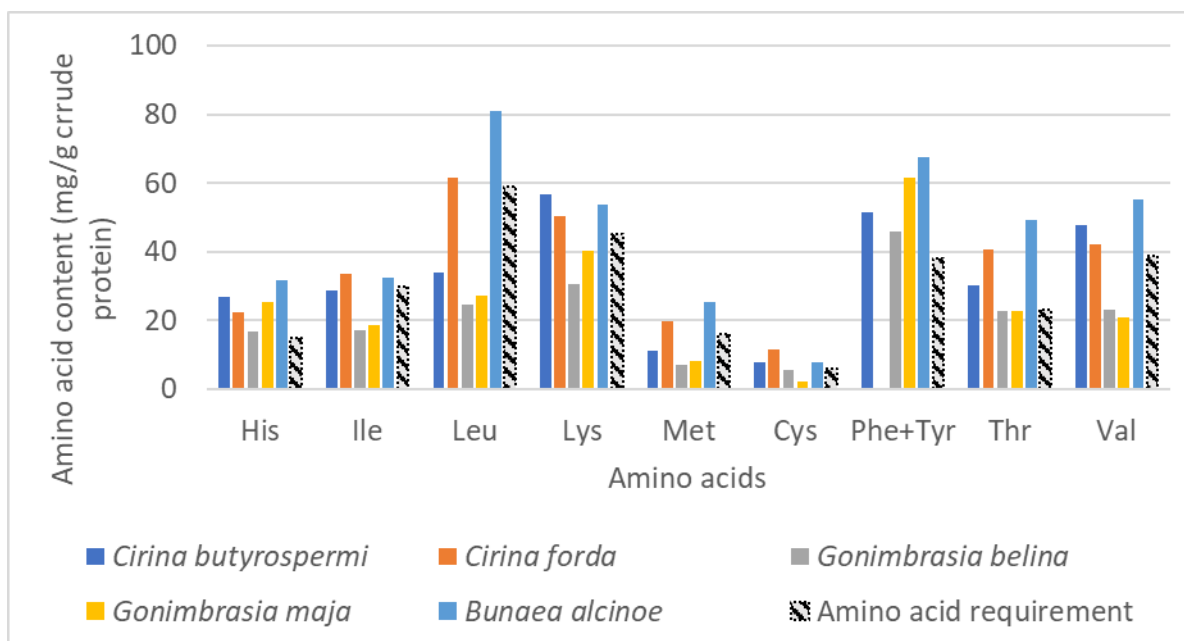


Figure 2. Mean essential amino acid contents [mg/g crude protein] of edible saturniids from literature compared to amino acid requirements for adults (mg/g protein) (WHO, 2007) His, histidine; Ile, isoleucine; Leu, leucine; Lys, lysine; Met, methionine; Cys, Cysteine; Phe, phenylalanine; Tyr, tyrosine; Thr, threonine; Val, valine

Edible saturniids are also rich in minerals, and *B. alcinoe* and *C. forda* are particularly high in phosphorus and potassium, while *G. belina* and *G. maja* are high in calcium and magnesium. However, except for *C. forda* this data is based on only one study (Appendix 5), highlighting the insufficient knowledge on the mineral content of edible saturniids in Africa.

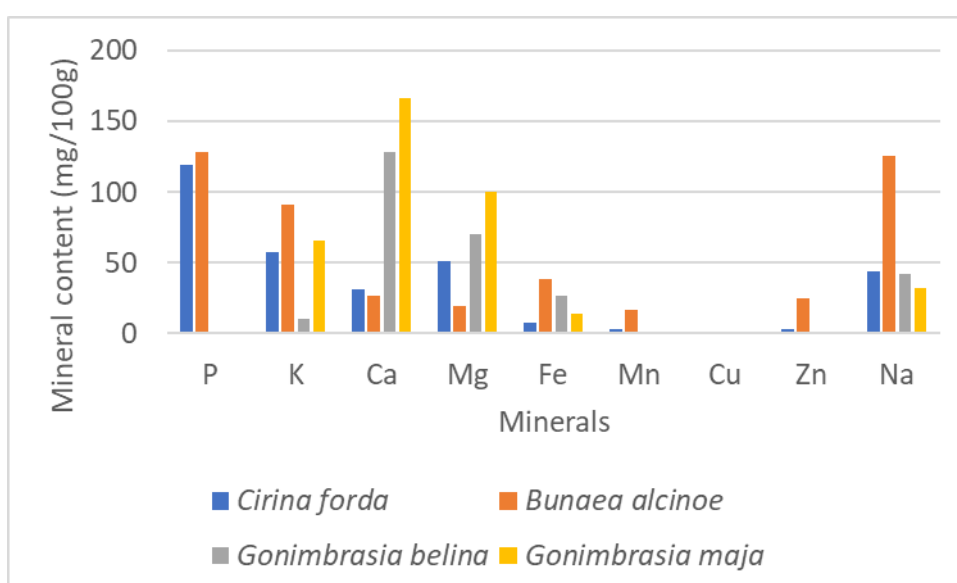


Figure 3. Mean mineral contents [mg/100g] of selected edible saturniids

Edible saturniids are to a varying degree rich in fats. For instance, linolenic acid (an essential fatty acid), stearic acid and palmitic acid are the most abundant fatty acids in *C. butyrospermi* (Anvo et al., 2016; Yapo et al., 2017). Oleic acid, stearic acid, and palmitic acid are the main fatty acids in *G. oyemensis* (Bi et al., 2015) while linolenic acid, stearic acid and oleic acids dominate the fatty acid profile of *C. forda* (Akinshaw and Ketiku, 2000), and linolenic acid, palmitic acid and oleic acid are the most abundant in *G. belina* (Glew et al., 1999).

2.11 Food safety

Edible saturniids are collected and processed in different ways depending on the region and community involved. Yet, these caterpillars may pose food safety concerns since they are harvested from the wild and hence it is difficult to set control measures. Traditionally, consumers use sight, taste and smell to determine whether the caterpillars are fresh. For instance, in Namibia, buyers either taste or watch out for mouldy caterpillars as a sign of poorly dried product (Thomas, 2013).

Processing (degutting, boiling/roasting and sun-drying) is expected to remove toxins, reduce post-harvest losses and aid preparing of the insect in a more palatable form (Agbidiye and Nongo, 2012). However, poor storage and poor sanitation when handling edible saturniid caterpillars may lead to fungal and bacterial contaminations. *Aspergillus* spp. has featured prominently as a fungal contaminant of mopane worms sourced from different markets in southern Africa. Among the fungi isolated are *A. flavus*, *A. parasiticus* and *A. ochraceus* which are well known producers of aflatoxins and ochratoxins. In Zambia, high concentrations of aflatoxin were found in sampled *G. maja* and *G. zambesina* (Kachapulula et al., 2018). Other toxigenic fungi detected in edible saturniid caterpillars in Africa included *Penicillium* spp. and *Fusarium* spp. (Mpuchane et al., 1996; Simpanya et al., 2000). In market bought and field collected mopane worms, *Escherichia coli* and *Klebsiella pneumoniae* contamination were identified, and *Enterobacteriaceae* were still present after cooking the caterpillars, indicating faecal contamination (Gashe et al., 1997). In ash roasted mopane caterpillars *Staphylococcus aureus* was found (Mujuru et al., 2014), as were *Aspergillus* spp., *Penicillium* spp. and *Fusarium* spp. in roasted and sun-dried *B.*

alcinoe larvae (Braide et al., 2011). In addition, bacteria including *E. coli*, *Pseudomonas aeruginosa*, *Proteus mirabilis* and *Streptococcus mitis* were also isolated from *B. alcinoe* samples (Braide et al., 2011).

In summary, poor sanitation during handling of edible saturniids combined with poor processing (delayed and uneven drying) and poor storage are the causes of bacterial and fungal contamination. Considering that edible saturniids are sold in open markets and as ready-to-eat snacks by street vendors, there is need to ensure improved hygiene practices during processing and handling of edible saturniids to reduce contamination.

2.12 Conservation concerns and interventions

Some of the major challenges affecting edible saturniids is over-harvesting and destruction of their natural habitats. Most saturniid host plants also have other uses including timber, firewood, fruit trees and medicinal uses (Ngute et al., 2020). Edible saturniids are most often harvested from the wild, underlining the importance to conserve their host plants.

Some African governments have started to put conservation measures in place. For instance, in Namibia there are restrictions on harvesting of mopane worms by giving permits to harvesters to curb over-exploitation. However, these permits do not specify the amounts and size of caterpillars that are allowed to be harvested (Thomas, 2013), and in addition illegal harvesting of mopane worms is still widespread. Monitoring is poor and offenders are rarely punished (Thomas, 2013). Establishing co-operative societies could be one strategy to better manage such resources (Thomas, 2013). Presently, authorities in Namibia lack sufficient rules governing the management and utilization of mopane worms since the insects are considered of low economic value. Most of the Namibian mopane belt is located in communal areas where informal customary laws permit anyone to harvest. However, some communities have set up local regulations to curb mopane worm over-exploitation. For example, in the Uukwaluudhi area a traditional authority was set up which regulates harvesting of mopane worms, announcing the availability of mopane caterpillars via radio and community gatherings. Illegal cutting of the mopane tree for firewood or building is prohibited, and locals are also discouraged from setting fires in the forest and over-harvesting caterpillars since this would jeopardize the subsequent development of mopane populations (Thomas, 2013). Apart from human interference, other factors

like prolonged drought and floods adversely affect mopane populations (Thomas, 2013)

Similarly, conservation concerns have been raised in Nigeria in relation to the shea tree caterpillar. Among the recommendations, forest conservation and establishing shea tree plantations to increase insect populations feature prominently. One of the interventions that is already yielding results is using pitfalls to harvest the caterpillars which prevents over-harvesting by avoiding breaking tree branches to reach caterpillars from the shea trees (Agbidye and Nongo, 2012). The Tiv community in Nigeria have developed traditional conservation strategies for *C. forda* which include delaying cultivation around *V. paradoxa* trees until adult moths emerge to reduce disturbance of the pupae buried in the ground. They also allow a few larvae to pupate during harvesting by sparing some host trees to ensure future generations of the insect. Some people even stop cultivation completely around *V. paradoxa* trees that were previously infested by *C. forda* (Agbidye and Nongo, 2012).

In Zambia, the Bisa community considers cutting trees that host edible caterpillars like *G. maja* and *G. belina* a taboo. However, population pressure and commercialization unfortunately weakened such rules with some locals ignoring informal rules and cutting tree branches to collect the caterpillars (Mbata and Chidumayo, 2003). In Zimbabwe, caterpillar populations decrease in December to January prompting harvesters to collect the insects in premature stages which leads to over-harvesting negatively affecting the subsequent development of insect populations (Kozanayi and Frost, 2002).

Traditionally, in the Bas Congo province of DRC, the first generation of *C. forda* is not harvested and caterpillars found high up on tree branches are spared. However, in many villages these safeguards are no longer adhered to. The other concern is burning bushes to clear land for cultivations and to repel rats; this kills the soil-dwelling saturniid pupae, hence greatly reducing caterpillar populations (Latham, 2015).

Logging also adversely affects edible saturniid populations. Edible saturniids are categorised as non-timber forest products. Cutting down high value timber trees like *E. cylindricum* has led to a decrease in saturniid caterpillars, as these trees usually have a wider crown hence carrying sizeable caterpillar populations (Muvatsi et al., 2018). While edible saturniids are an important resource, it is difficult to safeguard them in regions where logging for timber is a major activity, mainly for economic reasons. A good example is Sapelli (*Entandrophragma cylindricum* Harms, Meliaceae)

and Tali (*Erythropheum suaveolens* (Guill. & Perr.) Brenan, Fabaceae) trees in DRC that are important host plants for *G. oyemensis* and *C. forda*, respectively. Forest management there is often more geared towards safeguarding forests for logging and not necessarily for the sustainable harvesting of edible caterpillars (Karsenty and Ferron, 2017; Muvatsi et al., 2018). Trees, especially those close to the forest borders where locals traditionally harvest caterpillars, should be safeguarded for harvesting while logging should be confined more to the inner parts of the forest.

Another example is the African pearwood *B. toxisperma*, a wild host plant of *C. forda* in Cameroon. Apart from being a class A timber species, the tree also produces fruits from which an edible oil is extracted. Efforts are in place to domesticate *B. toxisperma* and *E. cylindricum* since they are categorised as vulnerable under the IUCN RedList of threatened species. One conservation strategy is incorporating such tree species into agroforestry systems like cocoa plantations as shown with mango and the safou tree *Dacryodes edulis* H.J. Lam (Burseraceae) in Cameroon (Ngute et al., 2020).

2.13 Conclusion

Apart from being a highly nutritious food source edible saturniids also provide substantial economic benefits to many households in SSA. However wild harvesting is increasingly becoming unsustainable. Their seasonal nature also limits their potential to profit the communities involved. Rearing methods need to be developed to ensure a more continuous supply and to help conserve their natural habitats. Additionally, more research is needed to develop better processing and storage methods that are faster, less tedious and improve food safety quality of the caterpillars. Development of insect-based food products should also be explored to process the caterpillars into more palatable forms with a longer shelf life. Gender mainstreaming should be considered when developing policies to sustainably maximize the benefits of edible saturniids in SSA. Value chains of edible saturniids need to be improved to establish possible points of intervention for maximum utilization of the food resource.

3.0 Exploring Community Knowledge, Perception and Practices of Entomophagy in Kenya

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3.1 Abstract

In recent years, community's perceptions and acceptance of edible insects as novel foods has gained momentum worldwide. Despite the importance of entomophagy in Kenya, little information exists on knowledge, perceptions and practices of edible insects among different communities. This study seeks to address this information gap by conducting surveys on edible insects in Kenya. Semi-structured questionnaires were used to collect information from 161 respondents in western, eastern, central and coastal Kenya. The studies revealed that major insect groups consumed in Kenya include termites (88%), grasshoppers (28%), saturniids (8.3%), crickets (6.8%), compost grubs (3%) and lake flies (1.5%). However, this varied with regions. For instance, saturniids caterpillars such as *Cirina forda*, *Bunaea alcinoe* and *Gonimbrasia zambesina* were consumed mainly by the Giriama community in Kilifi, Coastal Kenya. The Giriamas frequently consumed saturniids to complement their diet, rather than a tactic for survival. Insect consumption was significantly affected by age, occupation and gender but not by region or educational level. Children (92.3%) and women (98.6%) were prominently involved in wild harvesting and sale of edible insects. The most common edible insects observed in local markets included termites in western and saturniids in coastal Kenya. Although 98.8% of the respondents were familiar with edible saturniids, only 67.1% were willing consumers. While 73% of the respondents were willing to rear saturniids primarily for income, they cited lack of ready markets as one of the major challenges. There is an urgent need to create awareness and promote processing of these insects into more palatable forms that can be marketed and consumed as an alternative for alleviating food insecurity and malnutrition in the region.

Keywords: Community perceptions, Edible insects, Edible caterpillars, *Cirina forda*, Saturniidae

3.2 Introduction

The demand for food is expected to rise by at least 60% owing to a projected growth of the world population to 9.7 billion by the year 2050 (UN 2019). Currently, about 820 million people globally and over 256 million people in Africa suffer from hunger. In Eastern Africa, a third of the population is affected by hunger (FAO 2019). In addition to hunger, people also suffer from poor nutrition. According to World Health Organization (WHO), 150.8 million children under the age of five are stunted due to protein inadequacy (Development Initiatives 2018), especially in the developing countries (Ghosh et al. 2012).

Availability of conventional protein sources such as, poultry, eggs, fish, meat and dairy at affordable prices is limiting. Further, their production is exhausting on environmental resources, land, water (Doreau et al. 2012) and in many ways compete with other human food needs. Globally, the livestock sector accounts for 14.5% of total anthropogenic greenhouse gas (GHG) emissions (Gerber 2013). Hence, there is an urgent need for alternative sources of protein that are environmentally sustainable.

Edible insects are a sustainable alternative to conventional protein sources (Belluco et al. 2017; Patel et al. 2019). Production of edible insects requires less land and water and produces less ammonia and GHGs (Oonincx et al. 2010; Oonincx and de Boer 2012) compared to other protein sources. Edible insects are rich in proteins, fats, minerals, fibre and antioxidants (Anvo et al. 2016; Di Mattia et al. 2019; Patel et al. 2019; Tao and Li 2018). For instance, proteins account for >50% of the proximate composition of edible saturniid caterpillars such as *Bunaea alcinoe* (Braide et al. 2010), *Gynanisa maja*, *Gonimbrasia belina* (Musundire et al. 2016), *Cirina forda* (Igbabul et al. 2015) and *Cirina butyrospemi* (Yapo et al. 2017). Additionally, most edible saturniid caterpillars contain all the essential amino acids (Braide et al. 2010; Paiko et al. 2014; Siulapwa et al. 2014; Yapo et al. 2017).

Utilization of insects as an alternative protein source, is gaining relevance globally in recent years (Charlotte et al. 2015; Dobermann et al. 2017; Hongbété and Kindossi 2017; Kelemu et al. 2015; Melgar-Lalanne et al. 2019; Pali-Schöll et al. 2018; Payne et al. 2016). More than 2,100 insect species are consumed traditionally by over two billion people globally (Jongema 2017). Edible insects are a source of nutrition and livelihood for families in Asia and Africa (Badanaro et al. 2014; Baiyegunhi and

Oppong 2016; Makhado et al. 2014; Mbata et al. 2002; Nonaka 2009; Stack et al. 2003).

Over 470 species of edible insects are consumed in 54 African countries. Among the 470 species, 100 were recorded in East Africa (Kelemu et al. 2015). Three insect orders dominated the list, namely Lepidoptera, Orthoptera and Coleoptera. Among the Lepidopterans, species such as the mopane worm (*G. belina*) is widely consumed in South Africa, Zimbabwe, Malawi, Zambia and Botswana. This caterpillar is harvested from the wild, processed and sold in local, regional and international markets, providing income for the communities (Allotey et al. 1996; Baiyegunhi and Oppong 2016; Makhado et al. 2014; Stack et al. 2003). Similarly, the Shea butter caterpillar (*Cirina forda*) is the most popular edible saturniid consumed in Western, Central and Southern Africa. The caterpillar is highly nutritious and of economic importance as an edible insect, while it is also an important pest of the Shea butter tree (Adepoju and Daboh 2013; Badanaro et al. 2014; Dwomoh et al. 2010; Osasona and Olaofe 2010). However, there has been a decrease in the consumption of edible insects in Africa due to the loss of traditional knowledge associated with entomophagy (Dube et al. 2013; Looy et al. 2014; Obopile and Seeletso 2013; Riggi et al. 2016) and destruction of habitats associated with edible insects. Over several centuries, entomophagy has been a part of Africa food culture (Ayieko and Oriaro 2008; Münke-Svensden et al. 2016). However, in recent years, edible insect consumption is projected as 'rural food', 'practice associated with primitive people' and as a source of food during famine only, referred to as 'starvation food' (Looy et al. 2014). Such changes in community perception on entomophagy is also a major reason for decline in insect consumption in developing countries such as Kenya (Looy et al. 2014; Verbeke 2015). Hence to revive the healthy and traditional practices like entomophagy among younger and urban populations in developing countries, studies stress the need to document traditional practices and perceptions associated with entomophagy (Christensen et al. 2006; Riggi et al. 2016; van Huis et al. 2015).

In Kenya, community perceptions and consumption of lake flies has been studied in Western Kenya (Ayieko and Oriaro 2008). Nutritional composition of termites, crickets and ants consumed by Luo community in western Kenya has also been studied (Christensen et al. 2006). However, little is known on community perception and practices related to entomophagy in other parts of Kenya and other edible insect species such as Saturniids. Understanding these perceptions is important to inform

possible points of intervention in the bid to mainstream insect consumption in Kenya. This study, therefore, examined perceptions, knowledge and practices of communities engaged in entomophagy in Western, Central, Eastern and Coastal provinces of Kenya with a particular emphasis on edible saturniids.

3.3 Methods

3.3.1 Study area

This study was carried out between March 2017 and July 2017. The study sites are located in Western (Siaya, Homabay, Kakamega, Vihiga and Trans-Nzoia), Central (Meru, Nyeri, Laikipia and Embu), Eastern (Machakos, Makueni and Kitui) and Coastal Kenya (Taita, Kilifi and Kwale) (Figure 4). These regions are characterised by different cultures and ethnic groups with varied food cultures. In Western and Coastal Kenya, the culture of entomophagy is widespread, while Central and Eastern Kenya culture of entomophagy is not widespread. The study was carried out among rural communities.

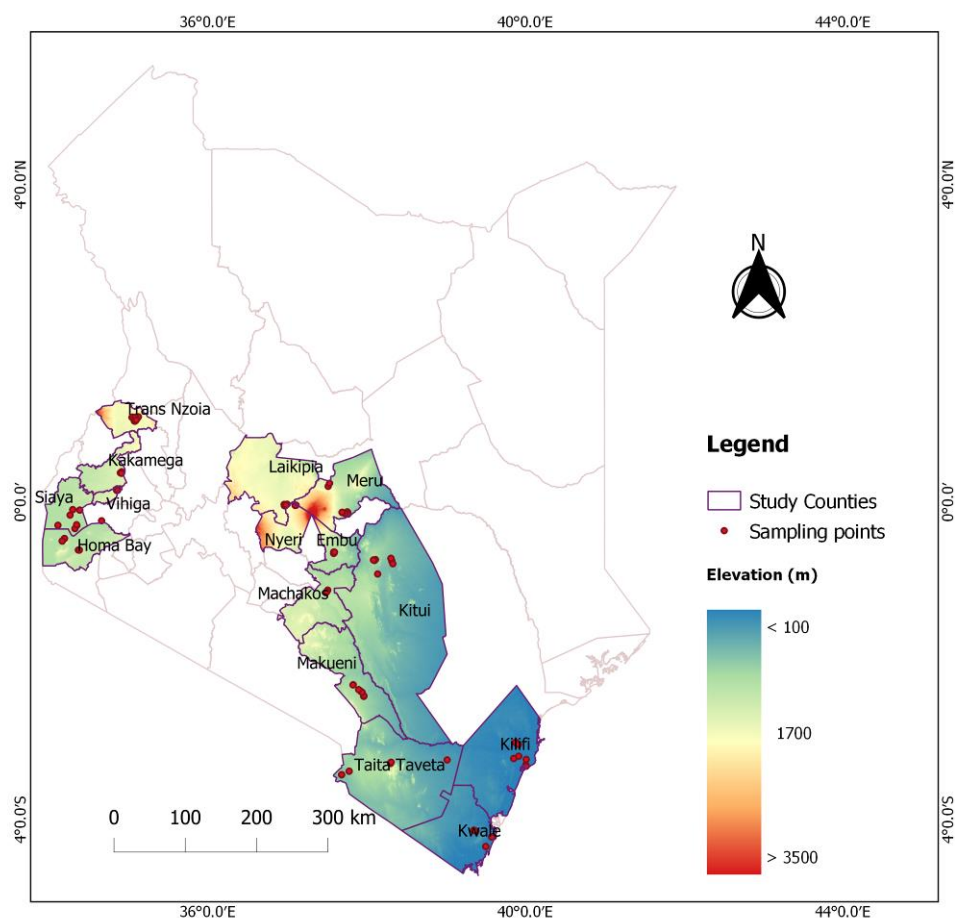


Figure 4 Map of Kenya showing study region and sampling sites

3.3.2 Study design and data collection

Semi-structured questionnaires were used to interview respondents who were chosen at random and had agreed to participate. A verbal description and colour pictures of edible insects were used during the interviews to help respondents identify edible insects. The pictures included six saturniid species (*Bunaea alcinoe*, *Cirina forda*, *Gonimbrasia zambesina*, *Gonimbrasia krucki*, *Gonimbrasia belina* and *Gynanisa nigra*), two species of grasshoppers (*Acanthacris* sp. and *Ruspolia* sp.), house cricket (*Acheta domesticus*) and termites (*Macrotermes* sp.). A total of 161 respondents were interviewed with a balanced mix of ages and gender (Table 1). They constituted 45 from Central region, 45 from Western region, 36 from the coastal region and 35 from the Eastern region. The questionnaire's structure began with questions on edible insects in general and narrowed down on edible saturniids. Data collected included 1) consumption, collection and trading of insects; 2) knowledge, consumption and perception of saturniids; 3) possibility and willingness to be involved in mass rearing saturniids; and 4) expected challenges/opportunities during rearing of saturniids.

In regions where saturniids are consumed, information on their collection, consumption, local names and host plants from which the insects are collected was recorded. Information on the potential for mass rearing of saturniids was collected from all the regions including regions where saturniids were not consumed.

Data was analysed using descriptive statistics in Microsoft Excel. Informants were segmented based on sex, gender, level of education and main economic activity. Insect consumption among different groups was analysed by means of Chi square test. For other comparisons between two groups, statistical significance was determined using t-test analysis. For situations where more than two groups were compared, Analysis of Variance (ANOVA) was utilized to test for statistical significance. This was followed by a Bonferroni corrected post hoc t-test to determine the significant differences between the groups.

3.4 Results

3.4.1 Socioeconomic background of the respondents

The respondents in this study were categorized into age, gender, educational level and main economic activity. Male respondents constituted 40.4% (n=65) while 59.6% (n=96) were female. Those aged below 30 years constituted 19.2% (n=31) of the respondents, while 43.5% (n=70) were between 30-50 years and 37.3% (n=60) above 50 years. Education level was classified as no formal education, primary school, secondary school and tertiary education. At 52.2% (n=84) most of the respondents had attained primary school education. Respondents with secondary school and tertiary institutions level education were 26.7% (n=43) and 8.7% (n=14), respectively. Those without formal education constituted 12.4% (n=20). The majority of the respondents (64%, n=103) were practicing farming while 29.2% (n=47), 6.2% (n=10) and 0.6% (n=1) were traders, civil servants and students, respectively (Table 1).

Table 1. Demographic characteristics of edible insect informants in Kenya.

Variables	Frequency (n=161)	Percentage
Age		
<30	31	19.2
30-50	70	43.5
>50	60	37.3
Gender		
Male	65	40.4
Female	96	59.6
Education Level		
No formal education	20	12.4
Primary School	84	52.2

Secondary school	43	26.7
Tertiary education	14	8.7
Main economic activity		
Farming	103	64
Trading	47	29.2
Civil service	10	6.2
Student	1	0.6
Region		
Central	45	28
Western	45	28
Eastern	35	21.7
Coastal	36	22.3

3.4.2 Insect consumption

This study documents six types of edible insects in Kenya, namely termites, grasshoppers, crickets, saturniids, lake flies and compost grubs, of which at least one type was consumed by 81% (n=131) of the respondents. Nineteen percent (n=30) were non-consumers ($p<0.05$). The most consumed insect in this study was termites (88%) while the least consumed was lake flies (1.5%). Consumption of termites was significantly higher than all the other insects in the study ($p<0.05$). Grasshoppers, saturniids, crickets, and compost grubs constituted 28%, 8.3%, 6.8% and 3% of insects consumed, respectively. There was a significant difference between consumption of grasshoppers, lake flies and compost grubs ($p<0.05$). There was no significant difference between the consumption of lake flies, crickets, compost grubs and saturniids ($p = 0.706$). Saturniids and compost grubs were eaten as larvae, while termites, grasshoppers, crickets and lake flies as adults (Table 2).

Table 2. Consumption rate of different insect species in Kenya

Insect group	Consumption (%)	Life stage consumed	Region of consumption
Termites	88	Adult	All sites sampled
Grasshoppers	28	Adult	All sites sampled
Saturniids	8.3	Larvae	Kilifi
Crickets	6.8	Adult	Kwale, Siaya, Homabay
Compost grubs	3	Larvae	Vihiga, Kakamega
Lakeflies	1.5	Adult	Siaya, Homabay

3.4.3 Role of age, gender, education, region and economic activity in insect consumption

Among respondents aged between 30 and 50 years, 81.4% (n=57) consumed at least one insect type while 90% (n=54) of those aged more than 50 years were insect consumers. Insect consumption among respondents below 30 years was significantly lower 64.5% (n=20) compared to the other age groups ($\chi^2 = 8.7554$, $p = 0.0126$) (Table 3).

There was a significant difference in insect consumption between female and male respondents at 84.4% (n=81) and 76.9% (n=50), respectively ($\chi^2 = 9.601$, $p = 0.0019$). Based on the level of education, respondents who lacked formal education had higher insect consumption rate at 90% (n=18). Insect consumers among respondents who had attained primary school, secondary school and tertiary education were 80.9% (n=68), 81.4% (n=35) and 71.4% (n=10), respectively ($\chi^2 = 1.9047$, $p = 0.5924$) (Table 3).

In terms of occupation, the significantly highest share in insect consumption was found among farmers 91.3% (n=94) ($\chi^2 = 37.3327$, $p < 0.05$), followed by traders at 74.5% (n=35) and civil servants at 20% (n=2).

The western region had the highest proportion of insect consumption at 88.9% (n=40) while eastern had the lowest rate at 74.3% (n=26). However, there was no significant difference between the rates of insect consumption in different regions ($\chi^2 = 2.908$, $p = 0.406$) (Table 3).

Table 3. Insect consumption based on age, gender, education level region and economic activity.

Variables	N	Consume % (n)	Do not consume
Age *			
<30	31	64.5 (20)	35.5 (11)
30-50	70	81.4 (57)	18.6 (13)
>50	60	90 (54)	10 (6)
$\chi^2 = 8.7554$ $p = 0.0126$			
Gender*			
Male	65	76.9 (50)	23.1 (15)
Female	96	84.4 (81)	15.6 (15)
$\chi^2 = 9.601$ $p = 0.0019$			
Education level			
No formal education	20	90 (18)	10 (2)
Primary School	84	80.9 (68)	19.1 (16)
Secondary school	43	81.4 (35)	18.6 (8)
Tertiary education	14	71.4 (10)	28.6 (4)
$\chi^2 = 1.9047$ $p = 0.5924$			
Main economic activity			
*			
Farming	103	91.3 (94)	8.7 (9)
Trading	47	74.5 (35)	25.5 (12)

Civil service	10	20 (2)	80 (8)
Student	1	0	100 (1)

$$\chi^2 = 37.3327 \quad p < 0.05$$

Region

Central	45	80 (36)	20 (9)
Western	45	88.9 (40)	11.1 (5)
Eastern	35	74.3 (26)	25.7 (9)
Coastal	36	80.6 (29)	19.4 (7)

$$\chi^2 = 2.908 \quad p = 0.406$$

*Significant difference within group

3.4.4 Role of gender in insect collection and trading

Though statistically insignificant slightly more female than male respondents reported to be involved in collection of insects for consumption at 48.8% (n=64) and 37.4% (n=49), ($p = 0.224$), respectively. Yet, 92.3% (n=121), ($p < 0.05$) of them reported children as the main collectors of edible insects (Figure 5).

At 43.5% (n=70), ($p = 0.52$) nearly half of the respondents had seen edible insects being sold in the local markets. Among them, 90% (n=63) ($p < 0.05$) reported seeing termites being sold, while 10% (n=7) had seen edible saturniids at the local market. Termites were mainly sold in western region, while saturniids were sold in the coastal region. Majority of the respondents at 98.6% (n=69), ($p < 0.05$) reported that the sellers were women, while slightly more men sold edible insects compared to children at 27% (n=19) and 17% (n=12) ($p = 0.553$), respectively (Figure 5).

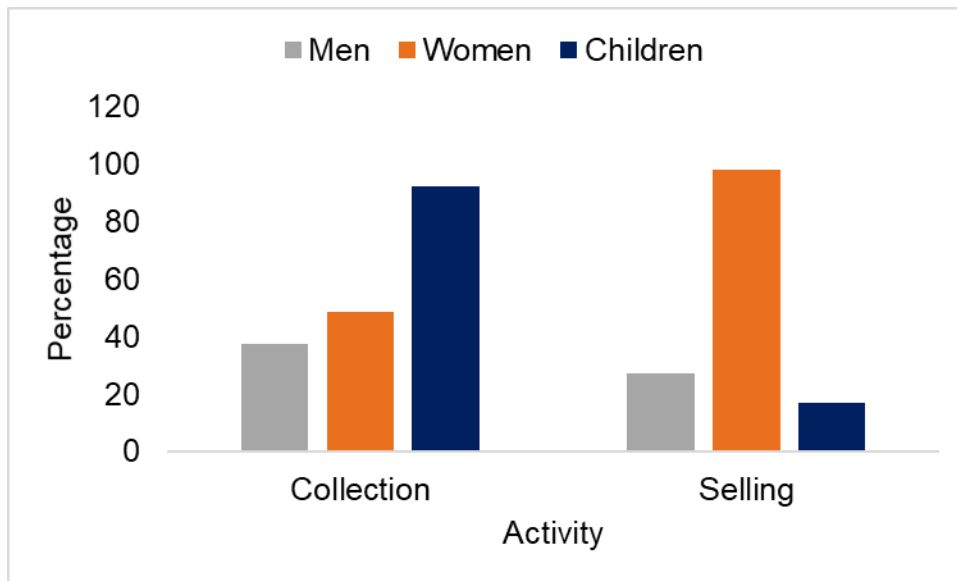


Figure 5 Participation of men, women and children in collection and selling edible insects

3.4.5 Perception of saturniids in Kenya

Respondents who had prior knowledge of saturniids were present in all the regions sampled and they constituted 98.8% (n=159) ($p<0.05$), while 1.2% (n=2) had never seen saturniids before. They reported that they had seen saturniid caterpillars during the rainy season. They had observed *B. alcinoe*, *C. forda*, *G. zambesina* and *G. krucki*. Among the respondents, 32.9% (n=53) had consumed or were willing to consume saturniids while a significantly larger proportion, 67.1% (n=108), ($p<0.05$) were not willing to consume them (Figure 6). Only respondents from the Giriama community consume saturniids in Kenya. Those who were not willing to consume saturniids 67.1% (n=108) were either scared of or disgusted by saturniids or considered them poisonous. The belief that saturniids could be harmful and poisonous was observed in all the regions sampled. Those willing to consume 32.9% (n=53) cited the need to process saturniid caterpillars into a more palatable form and suggested grinding them into powder and mixing with locally consumed flours. Respondents who had never consumed saturniids before sought to understand the benefits of consuming saturniids that could compel them to consume the insects. Generally, 77.6% (n=125) ($p<0.05$) perceived saturniids as harmful to humans, while 22.4% (n=36) and 20.5% (n=33) perceived them as crop pests and human food, respectively (Figure 6). There was no significant difference between the proportion that perceived saturniids as food and those that perceived them as crop pests ($p = 0.861$). In Meru, Eastern Kenya, some

respondents indicated that saturniid caterpillars were a sign of bumper harvests, while in Vihiga, Western Kenya, saturniid caterpillar appearance was a sign of the rains.

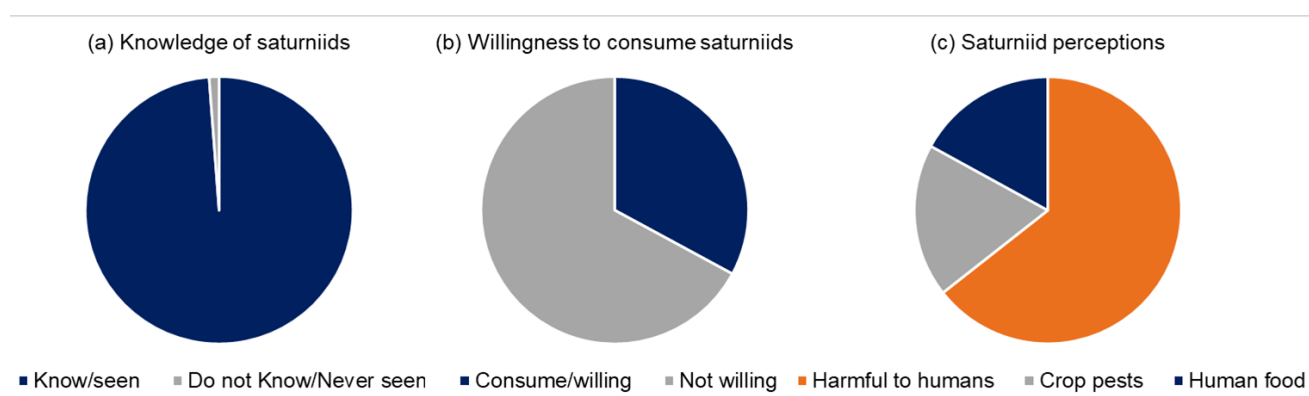


Figure 6 Pie charts showing knowledge, willingness to consume and general perception of saturniids

3.4.6 Consumption of saturniids in Kenya

Three saturniid species, namely *G. zambesina*, *B. alcinoe* and *C. forda*, were consumed in Kenya, and exclusively as larvae. Saturniids are widely consumed by the Giriama community in Kilifi County, with *C. forda* being the most popular and most abundant species. Saturniid caterpillars were generally referred to as '*maungu*'. They are combined with the name of the caterpillar's food plant to identify a particular species. For instance, *C. forda* fed on *Manilkara* sp. which is locally known as '*Msedzi*' hence the name '*maungu ya msedzi*'. On the other hand, *B. alcinoe* feeds on *Balanites* sp. locally known as '*mkonga*' hence '*maungu ya mkonga*'. *G. zambesina* is referred to as '*maungu ya mwembe*' since the larvae feed on the mango tree. Apart from consumption, the community also sold the surplus *C. forda* larvae in the villages and the nearby Malindi town whenever they were in season. Women and children were mostly involved in the collection, preparation and selling of *C. forda*. They collected the larvae by hand from the *Manilkara* trees and placed them in buckets. This was followed by squeezing out the gut contents by hand to avoid the leafy taste of the *Manilkara* plant. The insects were then cleaned, boiled in salty water and then spread out in the sun to dry. The dried insects were ready for long-term storage, immediate consumption or to be marketed. The dried larvae are consumed as such or fried with tomatoes and onions and accompanied by rice or the local maize meal called '*ugali*' (Figure 7).



Figure 7 (a) Dried *Cirina forda* for sale in Kilifi, Kenya; (b) *Cirina forda* fried in onions and tomatoes served with ugali

3.4.7 Potential of rearing edible saturniids, expected opportunities and challenges

Among the respondents, 78.9% (n=127) were involved in rearing insects. Among them, 95.3% (n=121) ($p<0.05$) reared bees, 2.4% (n=3) reared saturniids and 1.6% (n=2) reared butterflies and 0.7% (n=1) reared earthworms. Bee keeping was often for honey production, while saturniids (*G. zambesina* and *B. alcinoe*) and other butterflies were often reared on their respective host plants like *Mangifera indica* (mango) and *Balanites* sp. for later sale of the pupae to insect farms in Europe. With 73.3% (n=118) ($p<0.05$), a majority of the respondents were willing to rear saturniids. Among respondents who were willing to rear saturniids, income and consumption were cited as the potential opportunities by 94.9% (n=112) and 9.3% (n=11), ($p<0.05$) respectively. Potential challenges mentioned were lacking know-how of the rearing process, lack of ready markets and community perception at 97.4% (n=115), 34.7% (n=41) and 8.5% (n=10), respectively (Figure 8). Lack of know-how of the rearing process was cited by a significantly larger percentage of the respondents ($p<0.05$). Those who were not willing to rear saturniids were either scared of the caterpillars or not interested.

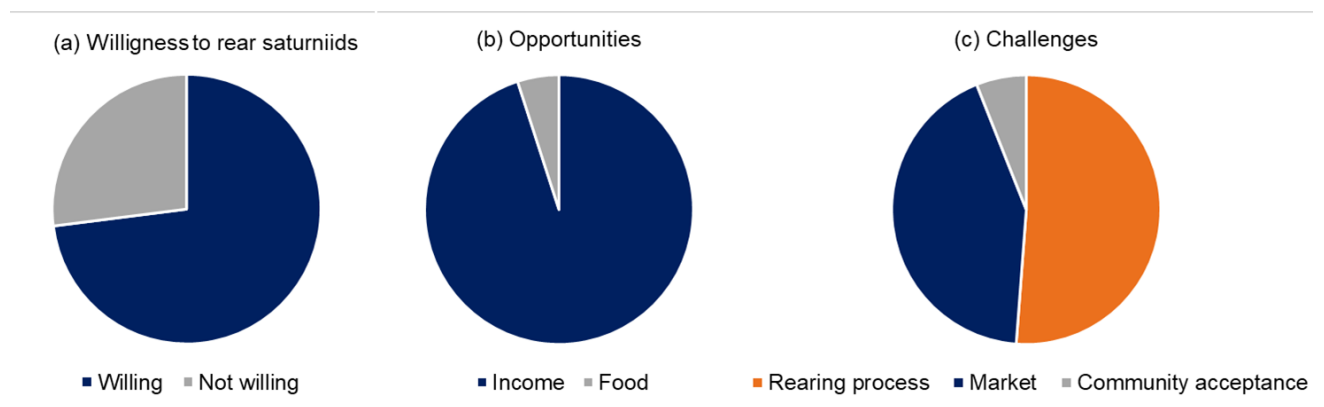


Figure 8 Pie charts showing willingness to rear saturniids and possible opportunities and challenges

3.5 Discussion

Consumption of insects has been practiced traditionally in different parts of the world. Consumption of termites, lake flies, grasshoppers, saturniids (*B. alcinoe*) and crickets has been previously reported in Kenya (Ayieko et al. 2010; Christensen et al. 2006; Münke-Svendsen et al. 2016; van Huis 2019). We report for the first-time consumption of saturniid caterpillars, *C. forda* and *G. zambesina* by the Giriama community in Kenya. These caterpillars are widely consumed in other parts of southern, western and Central Africa (Badanaro et al. 2014; Latham 2015; van Huis 2019). While lepidopterans are the most popular edible insect group in Africa (Kelemu et al. 2015), in Kenya, with 88% we found termites (*Isoptera*) to be most popular. The differences in consumption rates among different orders and species can be attributed to preference which is influenced by availability (Obopile and Seeletso 2013; van Huis 2013), ethnicity (Riggi et al. 2016) and seasonality (Kinyuru et al. 2013). The observed high prevalence of termite consumption could be associated with their availability within the study area. On the other hand, lake flies were the least consumed insect type as their distribution is limited to larger water bodies (Ayieko and Oriaro 2008).

Despite being present in most parts of Kenya, saturniids were consumed only by 8% of the respondents. This could be associated with culture and ethnicity since their consumption is confined within the Giriama community of Coastal Kenya. The people who did not consume insects were either scared of or disgusted by them. Previous studies have reported disgust as one of the reasons people shy away from consuming insects (Schardong et al. 2019). Food is cultural, hence cultural changes and modernisation can lead to changes in food-related practices. Although we report more

insect consumers than non-consumers, a reduction in the frequency of insect consumption in Kenya remains eminent. We reported lower insect consumption rate among respondents aged below 30 years than older ones. This is probably due to modernization among the youth, reduced edible insect populations and a knowledge gap between the young and older people regarding practices associated with insect consumption (Dube et al. 2013; Obopile and Seeletso 2013; Ramos-Elorduy 2009; Riggi et al. 2016). Hence, promoting entomophagy is still necessary, especially among the younger generations. However, in countries where entomophagy is still mainstream like Cote D'ivoire, more young people have been reported to consume insects more than their older counterparts (Ehounou et al. 2018).

Perception of insects as food for the poor (Tao and Li 2018) could also explain the reduction in insect consumption among the younger generations. Increasing agriculture is encroaching into the natural habitats of edible insects, which has also reduced their availability, leading to reduced consumption. Yet, in a study carried out in Ghana, age had no significant effect on insect consumption (Anankware et al. 2017). While 90% of respondents lacking formal education consumed insects, there was no significant association between education and insect consumption. A study carried out in Zimbabwe reported a negative association between education and insect consumption (Manditsera et al. 2018). There was a significant association between the main economic activity and insect consumption. More farmers consumed insects compared to civil servants probably because civil servants are closer to the city and adhere more to a westernized urban lifestyle compared to farmers.

More women consumed insects compared to men. Contrarily, in Brazil (Schardong et al. 2019) and Italy (Sogari et al. 2019), men were more inclined to insect consumption than women. A different trend was observed for the collection and sale of edible insects in the local markets. Insect collection was mostly carried out by children while selling was mostly left for women. Similar trends have been reported in southern Africa for the collections and sale of mopane worms (Stack et al. 2003). Contrarily, collection of *C. forda* in Togo is carried out by men, women and children in equal measure (Badanaro et al. 2014). In Kenya, men probably do not perceive insect collection and trading as a main source of income hence leaving it to women and children.

In some ethnicities (Meru and Luhya) in Kenya, occurrence of saturniid caterpillars was associated with the rain and a bumper harvest. Van Huis (2019) reported similar beliefs among the Luo community in Kenya and the Chaga, Sukuma and Digo in

Tanzania. While saturniid caterpillars are considered as food in some parts of Kenya, they are deemed harmful and poisonous in other parts, probably because of their conspicuous appearance. This points to the need for education and awareness creation to quell these beliefs and encourage consumption of saturniids.

Among the respondents, 32% were willing to consume saturniids. They suggested processing of the caterpillars to more palatable forms. A previous study in western Kenya showed that biscuits made of cricket flour were preferred compared to roasted whole crickets (Münke-Svendsen et al. 2016). In other parts of the world including Switzerland, Brazil, the United States of America and India, processed insects or foods containing insects were preferred compared to whole insects (Hartmann and Siegrist 2016, 2018; Ruby et al. 2015; Schardong et al. 2019). Processing edible saturniids into powders that could be included in other meals could promote their consumption. A study in Germany demonstrated that informing people about the benefits of consuming insects can increase consumption (Rumpold and Langen 2019). Increased awareness on the nutritional benefits of consuming edible saturniids could also increase consumption.

Most of the respondents who were willing to rear saturniids had been involved in bee keeping, rearing saturniids and butterflies. This implies that it is easier to introduce insect farming among communities that already practice insect farming. However, their main motivation for rearing saturniids was to generate income, while the main shortcoming was a ready market for their products.

3.6 Conclusions

Edible insects are widely consumed in Kenya. However, younger people consume less insects compared to older people. It is important to create awareness on the benefits of consuming insects and to develop more palatable foods using edible insects to promote consumption. Women and children play a crucial role in collecting and marketing edible insects. Hence strategies for establishing sustainable production and processing of edible insects should focus on the needs of women and could be an opportunity for women empowerment. Because of their widespread occurrence edible saturniids could be an important component of edible insects in Kenya, both in terms of local consumption and for development of edible insect-based products for market outside Kenya. Currently, saturniids consumption is confined to the coastal

region of the country, hence there is a need to create awareness and develop more palatable forms like powders and spread the practice of entomophagy further inland.

4.0 Diversity, Host Plants and Potential Distribution of Edible Saturniid Caterpillars in Kenya

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4.1 Abstract

Simple Summary: Edible insects are a traditional food source with economic benefits in sub-Saharan Africa. Caterpillars are the most popular edible insects in this region. We focus on caterpillars in the family Saturniidae. Saturniids are big colorful caterpillars with spines on their bodies, usually found in shrubs and trees. They are rich in proteins, vitamins, and minerals. Despite their economic importance, little is known about their diversity, host plants, distribution, and potential effect of climate change on edible saturniid caterpillars in Africa. The aim of this study is to identify edible saturniids, their host plants, their current distribution and to predict the possible effects of climate change on their distribution. We documented seven species of edible saturniids namely *Gonimbrasia zambesina*, *Gonimbrasia krucki*, *Bunaea alcinoe*, *Gonimbrasia cocaulti*, *Gonimbrasia belina*, *Gynanisa nigra* and *Cirina forda*. These caterpillars mostly occur twice a year during the rainy seasons and feed on specific host plants. Predictive distribution models revealed that *B. alcinoe*, and *C. forda* are mostly found in tropical and sub-tropical regions in Africa. However, climate change could cause a slight decrease in their population by the year 2050. This information will guide conservation efforts and ensure sustainable use of edible saturniid caterpillars as food.

Abstract: The promotion of edible insects, including saturniid caterpillars as potential food source is widely gaining momentum. They are adequately rich in nutrients such as proteins, amino acids, fatty acids, and micronutrients. Despite saturniids being a traditional food source with economic benefits, information on their diversity, host plants and their potential distribution in Africa are lacking, which this study seeks to address. Edible saturniids and their host plants were characterized using specific primers (LepF1/LepR1 and 3F_KIM_F/1R_KIM_R, respectively). Maximum entropy (MaxENT) and GARP (genetic algorithm for ruleset production) models were used to characterize the potential distribution of commonly consumed saturniids under current

and future climate scenarios. Seven species of saturniids were recorded from 11 host plants in Kenya: *Gonimbrasia zambesina*, *Gonimbrasia krucki*, *Bunaea alcinoe*, *Gonimbrasia cocaulti*, *Gonimbrasia belina*, *Gynanisa nigra* and *Cirina forda*. Two morphotypes of *G. zambesina* and *B. alcinoe* were recorded. These saturniid caterpillars occur twice a year except for *G. cocaulti*. Predictive models revealed that tropical and subtropical regions were potentially suitable for *B. alcinoe* and *C. forda*. The information generated from this study would be important to guide conservation efforts and their sustainable utilization as food in Africa.

Keywords: *Bunaea alcinoe*; *Cirina forda*; *Gonimbrasia zambesina*; *Gonimbrasia belina*; saturniids; edible insects; entomophagy; edible caterpillars; host plants

4.2 Introduction

The Food and Agriculture Organization of the United Nations has termed edible insects as one of the solutions to curb food insecurity (van Huis et al. 2013). Edible insects have been described as an alternative protein source (van Huis 2013) and recent studies have shown that they are a rich source of antioxidants (Di Mattia et al. 2019) and are beneficial to the human gut microbiota (Stull et al. 2018). Production of these insects is more sustainable compared to livestock since they require less land area (Oonincx and de Boer 2012), they offer a more efficient feed conversion (van Huis 2013) and emit less greenhouse gases (Oonincx et al. 2010). Furthermore, edible insects require less water for mass production (Miglietta et al. 2015) and have the potential to be reared on bio-waste (Dobermann et al. 2019; Magara et al. 2019), which could in turn lower the cost of production.

Globally, more than 2 billion people consume over 2000 species of edible insects (Jongema 2017; van Huis 2013). About 60% of edible insect species in Africa belong to the order Lepidoptera which includes edible saturniids (Kelemu et al. 2015). Saturniids occur widely across Africa, USA, Australia and Asia (Pinhey 1956, 1972, 1975). Edible saturniids availability and consumption has been documented in West (Ande 2002; Badanaro et al. 2014), Central (Latham 2015) and southern Africa (Baiyegunhi and Oppong 2016; Glew et al. 1999; Makhado et al. 2014). However, knowledge on the species diversity and edible saturniids consumed in East Africa is

scarce. Most studies on edible saturniid caterpillars concentrated on western and southern Africa (Hlongwane et al. 2020).

Edible saturniids are characterized by big larval forms with spines on the surface which pupate into cocoons (Scoble 1992) that are formed on the plants or leaf litters in the ground from which brightly colored moths emerge (Oberprieler 1995; Pinhey 1956, 1972, 1975). The most economically important saturniid that is consumed widely in southern Africa is the mopane worm, *Gonimbrasia belina* (Westwood). The larvae of *Go. belina* is an important food commodity among rural communities that live around the mopane woodland, i.e., the mopane belt across Angola, Zambia, Zimbabwe Mozambique, South Africa and Botswana (Stack et al. 2003). They have been commercialized and contribute considerably to the rural economies; for instance, in Limpopo, South Africa, 63% of the harvested worms are sold in the local markets (Baiyegunhi and Oppong 2016). Another edible saturniid larvae in Africa with high commercial value is the pallid emperor moth or shea defoliator, *Cirina forda* (Westwood). Its larvae are considered an important food source for many rural communities in Zimbabwe, Nigeria, Togo, Ghana, Zambia, D.R. Congo, Central African Republic, and South Africa (Badanaro et al., 2014; Stack et al., 2003; Fazoranti and Ajiboye, 1993).

Edible saturniid species have been observed to inhabit different bioecological zones with considerable seasonal variability, as well as having high specificity and preference to various host plants. For example, in southern and eastern Africa, they are known to occur in large outbreaks in arid and savannah regions (Pinhey 1956; 1975; Nyoka, 2003). Edible saturniids can either be bivoltine or univoltine depending on the region (Ande and Fazoranti 1997; Badanaro et al. 2014; Balinga et al. 2004; Dithogo 1996; Latham 2015; Oberprieler 1995). Larvae of the mopane worm have been observed to feed specifically on *Colophospermum mopane* Kirk ex J. Léonard (Fabaceae) (Moruakgomo 1996) while, *C. forda* show higher preference for the shea butter tree, *Vittelaria paradoxa* C.F. Gaertn (Sapotaceae) in West Africa (Ande and Fazoranti 1998; Dwomoh et al. 2010).

Edible saturniid caterpillars are highly nutritious, providing vital vitamins, lipids and proteins and microelements to households, especially women and children (Adepoju and Daboh 2013; Di Mattia et al. 2019; Glew et al. 1999; Omotoso 2006; Womeni et al. 2009). Despite being highly nutritious, the diversity of edible saturniid caterpillars has not been studied in Kenya. Semi-wild rearing of edible saturniid caterpillars could

promote proper land use management and forest conservation in an agroforestry setting (Vantomme et al. 2004). The scarcity of information on edible saturniids and their distribution and host plants in Kenya has hampered the prospects of promoting their sustainable access and consumption as food among communities in Kenya. Knowledge on the host plants and the distribution of saturniids in Kenya may also promote their conservation in the ecosystem. The information on the host plants could open new opportunities for their mass production to ensure continuous supply. Therefore, the aim of the present study was to establish the diversity, distribution, and host plants of edible saturniids in Kenya. Further habitat suitability maps were also generated for selected saturniid species to assess their distribution under present and future climate change scenarios.

4.3 Materials and Methods

4.3.1 Study area

The study was carried out between March 2017 and May 2019 across different agro-ecological zones (FAO 1996; Sombroek et al. 1982) and altitudes in Kenya. The highland areas (1200–2000 m above sea level (meters above sea level, m.a.s.l.)) included Nakuru, Laikipia, Tharaka Nithi, Embu, Meru, and Nairobi counties, while the lowlands areas (0–750 m.a.s.l.) were represented by the counties of Makueni, Taita, Kwale and Kilifi. The middle altitude areas (750–1200 m.a.s.l.) included Homabay, Kitui, Kajiado and Machakos counties (Table S1). Some additional samples collected from Ibadan, Nigeria and Tutume, Botswana alone were included for comparison with samples collected in Kenya.

4.3.2 Sample Collection and Preparation

Saturniid larvae were sampled at random along motorable roads from 15 sites (Appendix 6) in Kenya. Mostly fourth and fifth instar stages were collected, placed in buckets with twigs from their host plant and transported to *icipe*, Nairobi, Kenya, for rearing and identification. Twigs of host plants were collected and pressed in a herbarium for identification. Sampling was done for five rainy seasons between March 2017 and May 2019, both long (March to May) and short rains (October–December), in Kenya.

Field-collected Saturniid larvae were reared at 12 h:12 h photoperiod at 25 °C in Perspex cages (50 × 50 × 50 cm) with nets on the sides for ventilation. Cages were protected from ants and crawling insects with traps containing water placed below its metallic stand. The saturniid larvae were fed on twigs of their respective host plant from the field. The twigs were placed in a container as bouquet with stem immersed in water fastened by wet cotton wool. The twigs were changed daily to keep them fresh. The larvae fed until they reached the pre-pupal stage, when they stopped feeding, reduced movement, and moved to the floor of the cage ready to burrow and pupate. The pre-pupae were placed on moist sterile sawdust in plastic trays and allowed to burrow into the sawdust to pupate as they burrow in soil in the wild. The saw dust was kept moist by sprinkling water daily. The trays with the pupae were placed in Perspex cages (50 × 50 × 50 cm) awaiting emergence of adult moths.

Table 4. Morphological characteristics of saturniids collected in Kenya

Species	Characteristics			
	Moth Color	Hindwing Eyespot Description (Innermost to Outermost Color)	Mature Larvae Color	5th Instar Spine Color
<i>Gonimbrasia zambesina</i>	Green	Greenish-yellow center, black, greenish-yellow, white rings	Black with grey and yellow speckles	Black or red
<i>Gonimbrasia zambesina</i>	Brown	Yellowish brown center, black, pink, white rings	Black with grey and yellow speckles	Red
<i>Gonimbrasia cocaulti</i>	Brown	White center, reddish, black and white rings	Black with whitish speckles	Yellow
<i>Gonimbrasia krucki</i>	Yellow	Yellow center, black, pink, red rings	Black with greenish-yellow speckles and orange spots on spiracles	Black
<i>Cirina forda</i>	Light Brown	Small with a black ring	Black with yellow bands	White
<i>Gonimbrasia belina</i>	Reddish-Brown	Brown center, black, white rings	Black with red, grey and green speckles	Black
<i>Bunaea alcinoe</i>	Dark brown	Orange center, black, white rings	Black with orange spots on spiracles	White/yellow
<i>Bunaea alcinoe</i> *	-	-	Red	White
<i>Gynanisa nigra</i> *	-	-	Green with white speckles	White
<i>Gonimbrasia belina</i> *	-	-	Black with red, grey and greenish speckles	Black

* Adults moths were not collected

4.3.3 Morphological Identification

The adult moths were killed by placing them in a container and freezing them at -5°C . They were stretched out, pinned, allowed to dry, and labeled before morphological identification. Identification was carried out using published keys (Pinhey 1956) and crosschecked with reference voucher specimens at National Museums of Kenya (NMK) collection and pictures from available literature (Goff 2019; Kuhne 2008) by Mr. Alex Musyoki, Mr Ashikoye Okoko and Dr. Esther Kioko, NMK. Voucher specimens are deposited at the Biosystematics Unit, *icipe*. Host plants were identified at the Kenya Forest Research Institute (KEFRI) by an experienced plant taxonomist using available literature (Beentje et al. 1994).

4.3.4 Molecular identification

Tissue Preparation, DNA Extraction and Quantification

Leaf samples of each host plant of saturniid caterpillar were carefully washed with tap water, rinsed with distilled water and dried with paper towel. They were then cut into 0.2 g of leaf sample small pieces with a sterile blade and placed in a 2 mL tube containing ceramic beads, lysis buffer PA1 (Bioline, London, UK) and RNase A and crushed for 3 min in a Tissue lyser II (Qiagen, Germantown, MD, USA). Plant genomic DNA was extracted using Isolate II Plant DNA extraction Kit (Bioline, London, UK) as per the manufacturer's instructions.

A leg of each adult moth and/or a portion of larvae collected were cut with a sterile blade and placed in a 2 mL tube. Insect genomic DNA was extracted using Isolate II genomic DNA extraction kit (Bioline, London, UK) as per the manufacturer's protocol. The resultant DNA was eluted in 50 μL Elution buffer (Bioline, London, UK) and quantified using a NanoDrop 2000/2000 c spectrophotometer (Thermo Fisher Scientific, Wilmington, NC, USA). Insect and plant DNA samples were stored at -20°C for further downstream processing.

PCR for Insect Samples

For insect identification, PCR was conducted using general insect DNA barcoding LepF1/LepR1 primers (LEP F1-5' ATTCAACCAATCATAAAGATATTGG 3'; LEP R1 5' TAAACTTCTGGATGTCCAAAAAATCA 3') (Hebert et al. 2004). Isolated insect DNA was amplified in 30 μL PCR mix containing 17.025 μL PCR water, 6 μL My Taq Buffer (Bioline, London, UK), (5 mM dNTPs, 15 mM MgCl_2 , stabilizers and enhancers), 1.5

μL of each primer, 0.6 μL of 25 mM MgCl₂ (Thermo Fisher Scientific, Waltham, MA., USA), 0.375 μL 1 unit My Taq DNA polymerase (Bioline, London, UK) and 15 ng/L of DNA template. The reaction was set up in a Mastercycler Nexus Gradient thermocycler (Thermo Fisher Scientific, Waltham, MA, USA) using conditions as follows: initial denaturation at 95 °C for 2 min followed by 40 cycles of denaturation at 95 °C for 30 s, annealing at 52 °C for 40 s and primer elongation at 72 °C for 1 min. The final extension step lasted for 10 min at 72 °C.

PCR for Plant Samples

General plant primers 3F_KIM_F/1R_KIM_R (3F_KIM_F 5' CGTACAGTACTTTTGTGTTTACGAG 3'; 1R_KIM_R5' ACCCAGTCCATCTGGAAATCTTGGTTC 3') were used to amplify a 900 bp region of the matK gene for the identification of host plants. Protocols for PCR of plant samples were similar to the PCR protocol for insect samples above, except the annealing step which was done at 49 °C for 45 s.

Agarose Gel Electrophoresis, PCR Product Purification and Sequencing

Resolution of the PCR product was done with 1% agarose gel stained with ethidium bromide (10 mg/mL) at 80 volts for 1 h (Bio-Rad model 200/2-0 power supply, Bio-Rad laboratories Inc., Hercules, CA, USA). DNA bands were visualized using an ultraviolet transilluminator and photographed using the KETA GL imaging system software (Wealtec Corp., Sparks, NV, USA). The resultant PCR products for both the insects and the host plants were purified using QIAquick PCR purification kit (Qiagen, Hilden, Germany) and quantified with a NanoDrop 2000/2000 c spectrophotometer (Thermo Fisher Scientific, Wilmington, NC, USA) before being sent for bidirectional sequencing at Macrogen Inc. (Amsterdam, The Netherlands).

Sequence Analysis

Both plant and insect sequences were assembled and edited using Bioedit software v. 7.0.5.2 (Hall 1999). A consensus sequence generated from both the forward and reverse strand was queried on Basic Local Alignment Search Tool (BLAST) (Altschul et al. 1990) and Barcode Of Life Data system (BOLD) (Rantnasingham and Herbert 2007) to determine similarity with sequences in the database. The default Species level barcode records were used. The top published hit on Bold was used for

identification. Multiple sequence alignments were created on Clustal W (Thompson et al. 1994). Pairwise distances were generated using Mega X (Kumar et al. 2018). Sequences were submitted to the GenBank (<https://www.ncbi.nlm.nih.gov/WebSub/>) (accessed on 10 March 2020) (see Appendix 7).

4.3.5 Distribution Modelling

Two species of edible saturniids were selected for distribution modeling, i.e., *B. alcinoe* and *C. forda*, because of their popularity and economic importance in sub-Saharan Africa (SSA). *Cirina forda* and *B. alcinoe* are widely consumed (Kelemu et al. 2015; Latham 2015; Dauda et al. 2014; Paiko et al. 2014; Mabossy-Mobouna et al. 2016) and traded (Badanaro et al. 2014; Agbidye et al. 2009; Ehounou et al. 2018; Ngute et al. 2020) in western, central, eastern, and southern Africa.

Occurrence Data

The occurrence data (species name, GPS co-ordinates) for the two species was collected during field surveys in Kenya and from the Global Biodiversity Information Facility (GBIF). A total of 96 points (Figure 9a) were acquired (59 from field surveys and 37 from GBIF) for *B. alcinoe*, while the *C. forda* dataset comprised 70 points (57 from field surveys and 13 from GBIF) (Figure 9b).

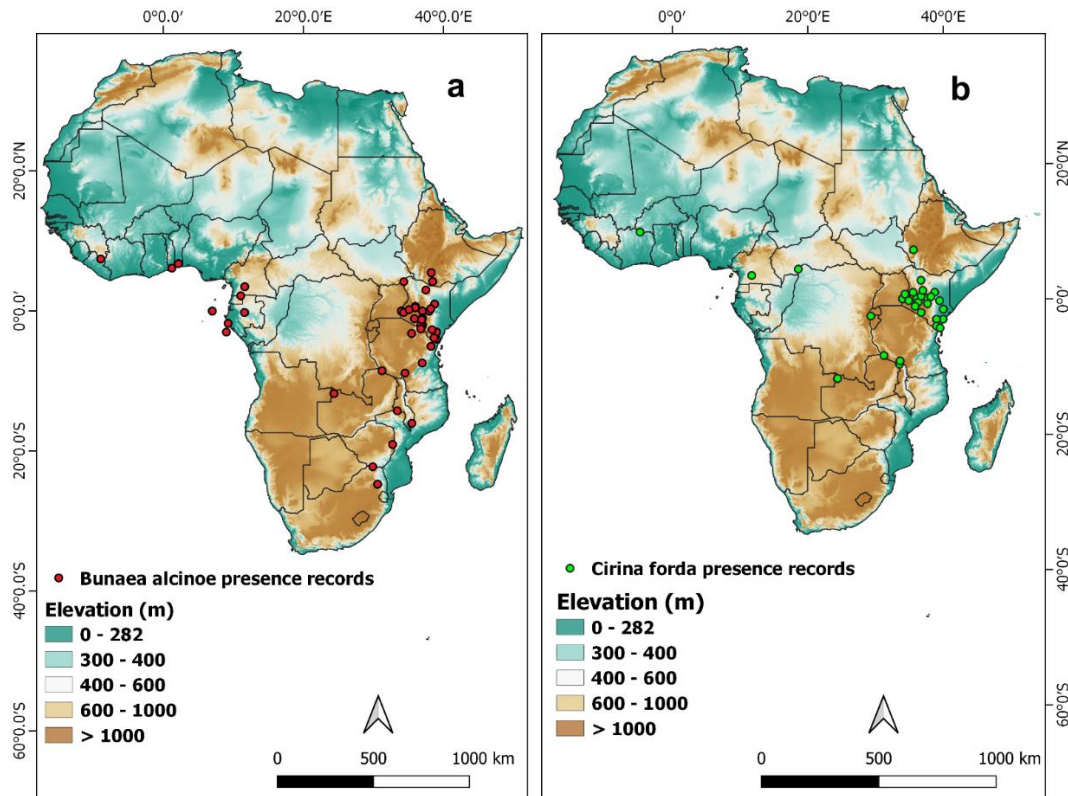


Figure 9. Geographical distribution records of (a) *Bunaea alcinoe* and (b) *Cirina forda* in Africa.

Environmental Variables

Nineteen bioclimatic variables downloaded from WorldClim (www.worldclim.org) (accessed on 15 April 2021) were considered for the study. We obtained current climate data for the period 1970–2000 at 30 arc seconds longitude/latitude degree spatial resolution (approximately 1 km at the equator). For future analysis, the downscaled and calibrated horizon 2050 IPPC (CMIP5) of climate projection bioclimatic variables were extracted from HadGEM2-ES global climate model (GCM) representing representative concentration pathways (RCP8.5) for future climate scenarios set by the intergovernmental panel on climate change (IPCC) [53]. RCP8.5 scenario predicts the mean global temperature increase projections of up to 3.7 °C. A collinearity test was conducted on the 19 bioclimatic variables to reduce collinearity between variables, to avoid overfitting of the model and variable inflation (Phillips et al. 2006). The variance inflation factor (VIF) test was used to assess the correlation between variables. The “vifcor” function in R software version 3.0.1 was used to run the VIF test (Phillips et al. 2006). Six

bioclimatic variables, namely Bio2 (mean diurnal temperature range), Bio3 (Isothermality), Bio5 (max temp of warmest month), Bio13 (precipitation of wettest month), Bio15 (precipitation seasonality), and Bio19 (precipitation of coldest quarter) were selected for the *B. alcinoe* species analysis, while seven bioclimatic variables namely, Bio2, Bio4 (temperature seasonality), Bio8 (mean temperature of wettest quarter), Bio13, Bio15, Bio18 (precipitation of warmest quarter), and Bio19 were selected for the *C. forda* species analysis using a cutoff of $|r^2| > 0.7$. Aside from the spatial correlation, the ecological relevance of the variables was also considered.

Model Calibration and Accuracy Assessment

Ecological niches of the two species were modeled using Maximum Entropy (MaxEnt) in the MaxEnt tool package version 3.4.1k which performs well for modeling presence only data (Phillips et al. 2006). The ENMEval package in R software was used to determine the required parameter settings to be used in Maxent software for the optimum tuning of the models (Muscarella et al. 2014). Following the parameter settings from ENMEvaluate, three features (linear, quadratic and hinge) were utilized with a regularization multiplier of 3. The model calibration created the optimal models for the two saturniid species. The models were replicated 3 times using cross-validation method and an ensemble of the three probability outputs were used to determine the optimum suitability and performance of the models. Seventy percent of the presence records were utilized to train the model while 30% of the points were used to validate the performance of the model. The comparative relevance of each environmental variable for the models of *C. forda* and *B. alcinoe* was evaluated using the overall percentage contribution, area under the curve (AUC), and the Jackknife test. AUC values of 0 indicate impossible occurrence area while 1 indicates optimal occurrence area. The ROC method has shown to be effective in evaluating model performance and being independent of prevalence (Swets 1988; Wei et al. 2018). Outputs of the models highlighting the intensity and extent of habitat suitability of the two species were mapped with values ranging from 0 (unsuitable) to 1 (optimum). Suitability levels were grouped into five categories as follows: very low (0–0.1), low (0.1–0.3), moderate (0.3–0.5), high (0.5–0.7), and very high (0.7–1).

4.4 Results

4.4.1 Morphological Identification of Edible Saturniids

Seven species of Saturniidae were identified in Kenya. They include *B. alcinoe*, *C. forda*, *Gonimbrasia zambesina* (Walker), *Go. cocaulti* Darge and Terral, *Go. belina* Westwood, *Go. krucki* (Hering) and *Gynanisa nigra* Bouvier. Dead larval stages of *Go. belina* collected from Botswana and a sample of *B. alcinoe* collected from Nigeria were included for comparison (Table 4).

Bunaea alcinoe

Larvae are black with orange spots on the spiracles along the sides of the body. One *B. alcinoe* larva collected from Nigeria is red in color. Larvae of both color forms of *B. alcinoe* have white/yellow spines (Figure 10). *Bunaea alcinoe* moths are dark brown in color with a large glass spot on the forewing (Table 4). The hind wing has an orange eyespot ringed with black followed by white (Figure 11).

Cirina forda

Larvae are black with yellow bands and white hairy spines. *Cirina forda* moths are smaller than the other moths. *Cirina forda* larvae in the same colony also presented in two color forms. Some had a black body with yellow bands while others had a black body with white bands (Figure 10). It is light brown in color with a small black eyespot on the hindwing (Table 4, Figures 10 and 11).

Gonimbrasia cocaulti

Larvae appear black with whitish speckles and yellow spines. *Cirina forda* moths are smaller than the other moths. It is light brown in color with a small black eyespot on the hindwing (Figure 11). *Gonimbrasia cocaulti* moths have a brownish ground color with eyespots on both the forewing and hindwing. The eyespot on the hindwing is white surrounded by reddish, black, and white rings while the one on the forewing is whitish circled by a brownish and white ring (Table 4, Figures 10 and 11).

Gonimbrasia krucki

Larvae are black with greenish-yellow speckles and black thick spines. *Gonimbrasia krucki* presented two color forms of their larvae. Larvae produced from the same egg clutch developed into forms that had a black body with either yellow or green speckles (Figure 10). *Gonimbrasia krucki* moths have a yellow ground color with defined eyespots on both the forewing and the hindwing. Both eyespots are yellow in color ringed with black, pink, and red (Figure 11).

Gonimbrasia belina

Larvae are red, grey, and green with black spines. *Gonimbrasia belina* moths are reddish brown in color with a brown eyespot on the hind wing circled by black and white rings. It has a small glass spot on the forewing. The front part of the hindwing is reddish in color (Figures 10 and 11).

Gonimbrasia zambesina

Larvae are black with yellow and grey speckles while some have black spines and others red spines (Table 4, Figure 10). *Gonimbrasia zambesina* moths occur in two color forms, green and brown. The green form is reddish purplish on the forward part of the hindwing. The eyespot on the hindwing is greenish yellow in the middle, circled by a black ring followed by greenish-yellow and white. The brown form has a yellowish-brown eyespot on the hind wing with black, pink, and whitish rings (Figure 11). The brown form is not available in the NMK collection. The dichotomous key (Pinhey 1956) reported the specimen as *Go. said* (Oberthuer). However, the author expressed uncertainty and suggested that it could be a form of *G. zambesina*. The green forms were collected from Kilifi, Embu and Kwale, while a mixture of the green and brown forms was collected from Makuyu in Murang'a County (all Kenyan sites). The green moths from Kwale and Kilifi produced larvae that were black with grey and yellow speckles and black spines (Figure 10). Green moths from Embu laid eggs that hatched into larvae that were black with grey and yellow speckles and with red spines. The green and brown moths were also observed to mate with each other. The brown and green moths collected in Makuyu mated among themselves (green and green/green and brown/brown and brown) to produce black larvae with grey and yellow speckles and with red spines (Figure 11).

Gynanisa nigra

Larvae of *Gy. nigra* are green with white speckles and white spines (Figure 10). We could not get adult moth from field collected *Gy. nigra* due to extensive parasitism.



Figure 10. Pictures of saturniid larvae. (a) *Gonimbrasia zambesina* larvae with red spines, (b) *Gonimbrasia zambesina* larvae with black spines, (c) *Gonimbrasia cocaulti*, (d) two color forms of *Gonimbrasia krucki* larvae; black with green speckles and black with yellow speckles, (e) *Cirina forda* larva black in color with yellow spots, (f) *Cirina forda* larva black in color with white spots, (g) *Bunaea alcinoe* black form observed in East Africa, (h) *Bunaea alcinoe* red form observed in West Africa, (i) *Gynanisa nigra* larvae, (j) *Gonimbrasia belina* collected in Kenya.



Figure 11. Pictures of saturniid moths. (a) *Bunaea alcinoe*, (b) *Cirina forda*, (c) *Gonimbrasia krucki*, (d) *Gonimbrasia belina*, (e) green form of *Gonimbrasia zambesina*, (f) brown form of *Gonimbrasia zambesina*, (g) mating pair of brown and green form of *G. zambesina*.

4.4.2 Molecular Identification

All the saturniid species showed 98.22–100% similarity to sequences in BOLD. *Gonimbrasia belina* collected in Kenya and *Gy. nigra* were 100% similar to sequence GBMNC60703-20 (*Go. belina*; BIN-BOLD:AAB6786) and GBMNC60687-20 (*Gy. Nigra*; BIN- BOLD:AED6623), respectively. *Gonimbrasia zambesina* sequences were 99.09–100% similar to SAPBA773-07 (*Go. Zambesina*; BIN-BOLD:AAD1339)) while

Go. krucki was 98.22–100% similar to SAPBA635-07 (*Go. Krucki*; BIN-BOLD:AAD8374)). *Cirina forda* had 99.38–99.85% similarity to SATWA281-07 (*C. forda*; BIN-BOLD:AAB6982) and STBOB620-08 (*C. forda*; BIN-BOLD:AAB6982). *Gonimbrasia cocaulti* sequences were >98% similar to SPBIS152-09 (*Go. Cocaulti*; BIN-BOLD:AEH8028). *Bunaea alcinoe* sequences from samples collected in Kenya were 98.78–100% similar to LSAFR2238-12 from South Africa (*B. alcinoe*; BIN-BOLD:AAA6757) (Table 5).

Gynanisa westwoodi and *G. belina* collected from Botswana were both >99% similar to STBOA580-07 (BIN-BOLD: ABY462) and SATWA003-06 (BIN-BOLD: AAB6786), respectively. *Bunaea alcinoe* sequences from sample collected in Nigeria was 100% similar to SATWA891-07 (*B. alcinoe*; BIN-BOLD: AAA6756) (Table 5).

Table 5. Identities of saturniids based on similarities with sequences from BOLD.

Collection Site	Sample Code	Species	% Similarity to BOLD Sequences	ID of Similar Sequences- Default BOLD Query (Collection Site)
Mwingi, Kenya	S30	<i>Gonimbrasia cocaulti</i>	98.88	SPBIS152-09 (Kenya)
Taita, Kenya	S34	<i>Gonimbrasia cocaulti</i>	98.88	SPBIS152-09 (Kenya)
Matuu, Kenya	S22	<i>Gonimbrasia cocaulti</i>	98.72	SPBIS152-09 (Kenya)
Taita, Kenya	S32	<i>Gonimbrasia cocaulti</i>	98.87	SPBIS152-09 (Kenya)
Taita, Kenya	S33	<i>Gonimbrasia cocaulti</i>	98.88	SPBIS152-09 (Kenya)
Matuu, Kenya	S28	<i>Gonimbrasia cocaulti</i>	98.72	SPBIS152-09 (Kenya)
Muhaka, Kenya	S48	<i>Gonimbrasia belina</i>	100	GBMNC60703-20 (Kenya)
Ibadan, Nigeria	Nigeria-1	<i>Bunaea alcinoe</i>	100	SATWA891-07 (Burkina-Faso)
Matuu, Kenya	S84	<i>Bunaea alcinoe</i>	99.24	LSAFR2238-12 (South Africa)
Matuu, Kenya	S85	<i>Bunaea alcinoe</i>	99.39	LSAFR2238-12 (South Africa)
Nanyuki, Kenya	S87	<i>Bunaea alcinoe</i>	99.24	LSAFR2238-12 (South Africa)
Nanyuki, Kenya	S88	<i>Bunaea alcinoe</i>	99.24	LSAFR2238-12 (South Africa)
Mbita, Kenya	S90	<i>Bunaea alcinoe</i>	99.39	LSAFR2238-12 (South Africa)
Mbita, Kenya	S91	<i>Bunaea alcinoe</i>	99.08	LSAFR2238-12 (South Africa)

Embu, Kenya	S92	<i>Bunaea alcinoe</i>	99.23	LSAFR2238-12 (South Africa)
Embu, Kenya	S93	<i>Bunaea alcinoe</i>	98.78	LSAFR2238-12 (South Africa)
Nairobi, Kenya	S2	<i>Gonimbrasia krucki</i>	100	SAPBA635-07 (Kenya)
Nairobi, Kenya	S3	<i>Gonimbrasia krucki</i>	100	SAPBA635-07 (Kenya)
Mbita, Kenya	S6	<i>Cirina forda</i>	99.54	SATWA281-07 (Cameroon)
Mbita, Kenya	S7	<i>Cirina forda</i>	99.54	SATWA281-07 (Cameroon)
Kilifi, Kenya	S55	<i>Cirina forda</i>	99.41	STBOB620-08 (Malawi)
Ngong, Kenya	2CF	<i>Cirina forda</i>	99.38	STBOB620-08 (Malawi)
Ngong, Kenya	5CF	<i>Cirina forda</i>	99.69	STBOB620-08 (Malawi)
Gilgil, Kenya	S54	<i>Cirina forda</i>	99.85	STBOB620-08 (Malawi)
Kilifi, Kenya	S17	<i>Gonimbrasia zambesina</i>	100	SAPBA773-07 (Kenya)
Kilifi, Kenya	S18	<i>Gonimbrasia zambesina</i>	100	SAPBA773-07 (Kenya)
Kambiti, Kenya	S79	<i>Gonimbrasia zambesina</i>	99.54	SAPBA773-07 (Kenya)
Kambiti, Kenya	S94	<i>Gonimbrasia zambesina</i>	99.39	SAPBA773-07 (Kenya)
Embu, Kenya	S95	<i>Gonimbrasia zambesina</i>	99.39	SAPBA773-07 (Kenya)
Embu, Kenya	S96	<i>Gonimbrasia zambesina</i>	99.58	SAPBA773-07 (Kenya)
Makuyu, Kenya	S97 Brown	<i>Gonimbrasia zambesina</i>	99.54	SAPBA773-07 (Kenya)

Makuyu, Kenya	S98 Brown	<i>Gonimbrasia zambesina</i>	99.54	SAPBA773-07 (Kenya)
Makuyu, Kenya	S99 Green	<i>Gonimbrasia zambesina</i>	99.54	SAPBA773-07 (Kenya)
Makuyu, Kenya	S100 Green	<i>Gonimbrasia zambesina</i>	99.09	SAPBA773-07 (Kenya)
Makuyu, Kenya	S101 Green	<i>Gonimbrasia zambesina</i>	99.39	SAPBA773-07 (Kenya)
Makuyu, Kenya	S102 Green	<i>Gonimbrasia zambesina</i>	99.39	SAPBA773-07 (Kenya)
Makuyu, Kenya	S104 Brown	<i>Gonimbrasia zambesina</i>	99.39	SAPBA773-07 (Kenya)
Makuyu, Kenya	S105 Brown	<i>Gonimbrasia zambesina</i>	99.54	SAPBA773-07 (Kenya)
Makuyu, Kenya	S107 Brown	<i>Gonimbrasia zambesina</i>	99.24	SAPBA773-07 (Kenya)
Botswana	IBB-1	<i>Gonimbrasia belina</i>	99.84	SATWA003-06 (Zambia)
Botswana	IBB-2	<i>Gonimbrasia belina</i>	99.85	SATWA003-06 (Zambia)
Kenya	GMB-2	<i>Gynanisa nigra</i>	100	GBMNC60687-20 (Botswana)
Botswana	GM-1	<i>Gynanisa westwoodi</i>	99.69	STBOA580-07 (Kenya)
Botswana	GM-2	<i>Gynanisa westwoodi</i>	99.69	STBOA580-07 (Kenya)

Gonimbrasia krucki and *B. alcinoe*-Nigeria species had a 100% similarity to sequences from BOLD database. *Gonimbrasia zambesina* had a within species pairwise distance range of 0–0.61 and a range of 0–0.91 between species and BOLD sequence SAPBA773-07 (BOLD:AAD1339) (Table 6). *Gonimbrasia belina* collected in Botswana and *Gy. westwoodi* had a 100% similarity within species, while they had a pairwise distance range of 0.15–0.16 and 0.3 with BOLD sequences SATWA003-06 (BOLD:AAB6786) and STBOA580-07 (BOLD:ABY4629), respectively (Table 6). *Gonimbrasia belina* collected in Kenya was 100% similar to the BOLD sequence, while *G. nigra* had a pairwise distance of 0.61. *Cirina forda* showed a pairwise distance range of 0.3–2.13 within species and 0.46–2.13 between species and BOLD sequence SATWA281-07 (BOLD:AAB6982) (Table 6).

Table 6. Genetic p-distance comparisons for saturniid species.

Insect Species (Sample Size)	Genetic p-Distance Range within Sample Species	BOLD Sequence Used for Comparison (BOLD BIN Cluster Number)	Genetic p-Distance Range between Sample Species and BOLD Sequence
<i>Gonimbrasia zambesina</i> (15)	0-0.61	SAPBA773-07 (BOLD:AAD1339)	0–0.91
<i>Gonimbrasia krucki</i> (2)	0.0	SAPBA635-07 (BOLD:AAD8374)	0.0
<i>Gonimbrasia belina</i> -Kenya (1)	0.0	GBMNC60703-20 (BOLD:AAB6786)	0.0
<i>Gonimbrasia cocaulti</i> (7)	0–1.52	SPBIS152-09 (BOLD:AEH8028)	1.42–2.88
<i>Gonimbrasia belina</i> -Botswana (2)	0.0	SATWA003-06 (BOLD:AAB6786)	0.15–0.16
<i>Cirina forda</i> (4)	0.3–2.13	SATWA281-07 (BOLD:AAB6982)	0.46–2.13
<i>Bunaea alcinoe</i> -Kenya (8)	0–1.52	LSAFR2238-12 (BOLD:AAA6757)	0.61–1.98
<i>Bunaea alcinoe</i> -Nigeria (1)	0.0	SATWA891-07 (BOLD:AAA6756)	0.0
<i>Gynanisa nigra</i> (1)	0.0	STBOC836-08 (BOLD:AED6623)	0.61
<i>Gynanisa westwoodi</i> (2)	0.0	STBOA580-07 (BOLD:ABY4629)	0.3

4.4.3 Molecular Differences among the Color Forms of *Gonimbrasia zambesina* and *Bunaea alcinoe*

Although *Go. zambesina* larvae and moths depicted different color forms, they identified as the same species using molecular characteristics. The genetic distance between all the *Go. zambesina* samples and the reference *Go. zambesina* sequence from BOLD (SAPBA773-07; BIN BOLD:AAD1339) was 0–0.61% while the genetic distance between all the samples and *Go. zambesina* sequence from BOLD (SAPBA772-07) was 1.07–1.23%. The genetic distance between the samples was 0–0.61%. All the samples included in the analysis had 658 bp (Appendix 8, Figure 12). The morphological difference of the two larvae color forms of *B. alcinoe* was also supported by molecular characterization. The red form with white spines (Nigeria-1) was 100% similar to SATWA891-07-COI-5P (*B. alcinoe*; BIN BOLD:AAA6756) from Burkina Faso. However, the same BOLD sequence had a 3.28–3.76% genetic distance from all the other black larvae forms of *B. alcinoe* collected from Kenya (see Table S4. All the other black forms with white spines from Kenya showed a genetic distance of 0.61–0.76% from LSAFR2238-12 (*B. alcinoe*; BIN BOLD:AAA6757) from South Africa. The same BOLD sequence had a genetic distance of 3.76% from the red color form collected from Nigeria. All the black forms clustered together with LSAFR2238-12 (*B. alcinoe*) while the red form clustered with SATWA891-07-COI-5P (*B. alcinoe*). All the samples included had 658 bp (Appendix 9, Figure 13).

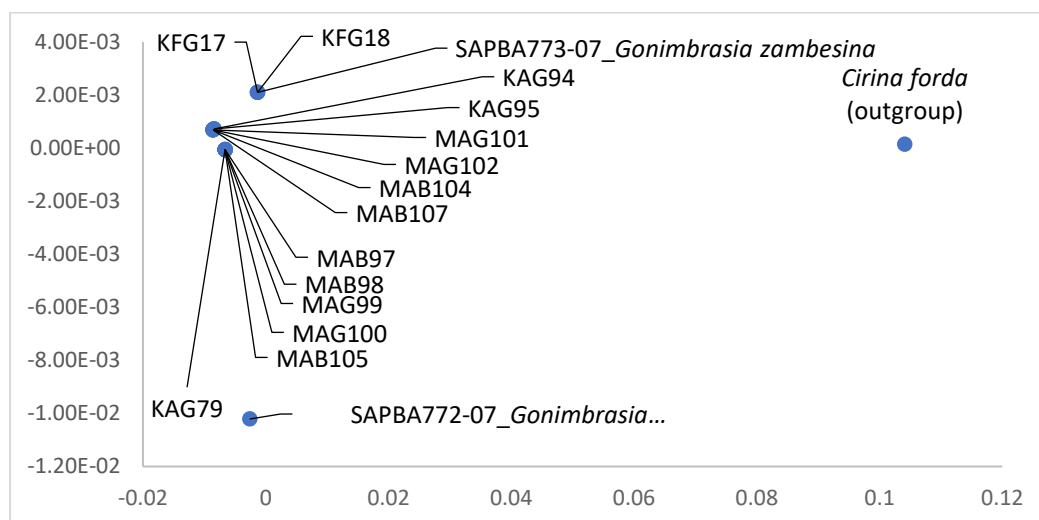


Figure 12. Principal coordinate analysis of COI gene of different morphotypes of *Gonimbrasia zambesina* from different locations in Kenya. The first two letters indicate

the collection sites (MA = Makuyu, KA = Kambiti, and KF = Kilifi). The last letter indicates the color of the adult moth (G = green and B = brown). The number indicates sample ID. SAPBA772-07 and SAPBA773-07 are BOLD reference accession numbers in BOLD BIN cluster BOLD:AAD1339.

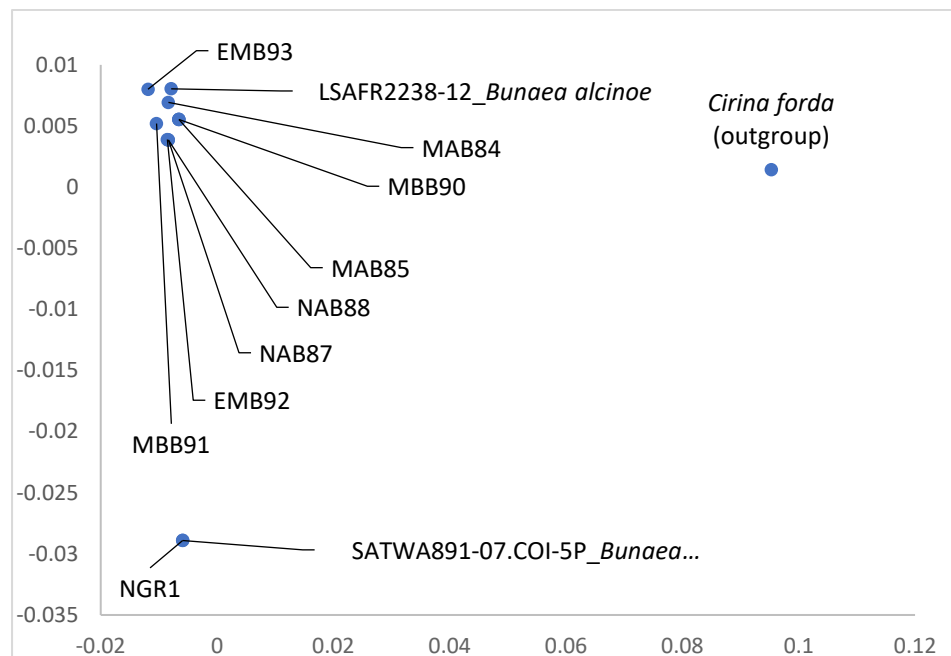


Figure 13. The principal coordinate analysis of COI gene of morphotypes of *Bunaea alcinoe* from Kenya and Nigeria. The first two letters indicate the collection sites (NG = Nigeria, MB = Mbita, EM = Embu, NA = Nanyuki and MA = Matuu). The last letter depicts the color of the larvae (R = red and B = black). The number indicates sample ID. SATWA891-07.COI-5P (BIN BOLD:AAA6756) and LSAFR2238-12 (BIN BOLD:AAA6757) are BOLD reference accession numbers.

4.4.4 Distribution and Seasonality of Edible Saturniids in Kenya

The distribution of the edible saturniids in the various Counties in Kenyan is presented in Appendix 6. *Gonimbrasia zambesina*, *C. forda*, *Go. krucki*, and *B. alcinoe* were bivoltine occurring between April–June and October–December, reflecting the major and minor rainy seasons in the region. On the other hand, *Go. cocaulti* was univoltine and occurred only during the April–June season (Table 7). The distribution of these saturniids was attributed to the availability of their host plants. The most widespread saturniid was *B. alcinoe* while the least widespread were *Go. belina* and *Go. krucki* which were only found in Kwale and Nairobi, respectively (Table 7).

Table 7. Seasonality and distribution of edible saturniids in Kenya.

Saturniid	Place Found	April– June	October– December
<i>Gonimbrasia zambesina</i>	Kilifi, Embu, Machakos, Kwale, Murang'a	Present	Present
<i>Cirina forda</i>	Kilifi, Nakuru, Embu, HomaBay, Kajiado	Present	Present
<i>Gonimbrasia cocaulti</i>	Taita, Makueni, Machakos, Kitui, Isiolo	Present	Absent
<i>Bunaea alcinoe</i>	Machakos, Makueni, Homabay, Meru, Kitui, Embu, Laikipia	Present	Present
<i>Gonimbrasia krucki</i>	Nairobi	Present	Present
<i>Gonimbrasia belina</i>	Kwale	Present	Present

4.4.5 Habitat Suitability and Probability Distribution

Area under Curve (AUC) Values

All the models using the current and future (RCP8.5:2050) showed a balance between goodness-of-fit and complexity (AUC > 0.80), for all the test and training datasets (Table 8). This demonstrates that our models showed good predictive performance.

Table 8. Area under curve values for training and test data.

Species	Current		RCP8.5	
	Training	Test	Training	Test
<i>Bunaea alcinoe</i>	0.855	0.915	0.877	0.928
<i>Cirina forda</i>	0.850	0.867	0.876	0.860

Visualization of Habitat Suitability under Current and Future Climatic Conditions

Habitat suitability maps show that the tropics are optimal for both *B. alcinoe* (Figure 14) and *C. forda* (Figure 15). Parts of the subtropical region in southern Africa are marginally suitable for *B. alcinoe*. For both MaxENT and GARP maps, a slight reduction in habitat suitability for both saturniids is predicted in future climate

scenarios. Northern Africa is unsuitable in both present and future scenarios for *B. alcinoe* and *C. forda*.

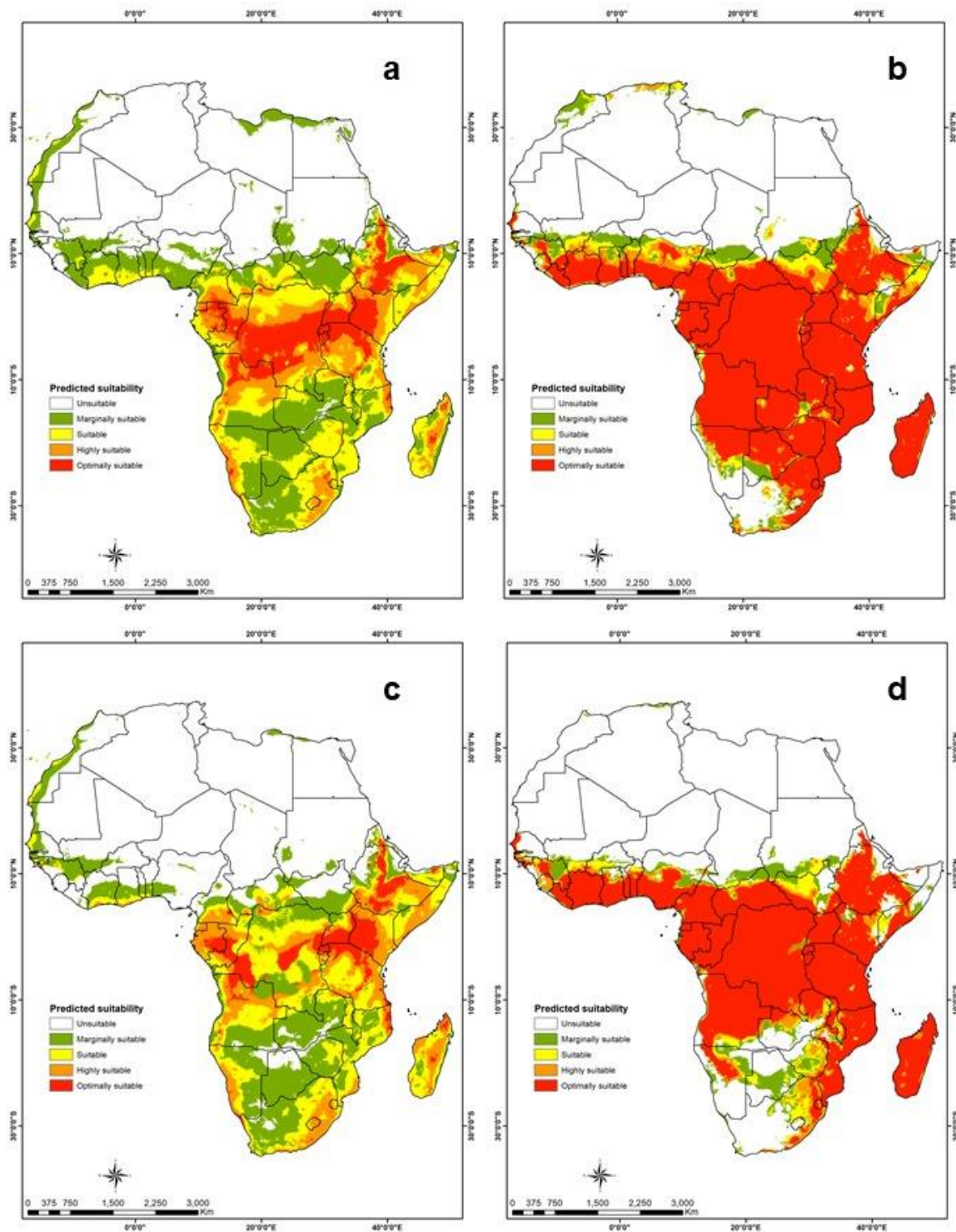


Figure 14. Predictive suitability map of the geographic distribution of *Bunaea alcinoe*. (a) Suitability map generated by the MaxEnt algorithm under current climate scenario; (b) suitability map generated by the GARP algorithm under current climatic scenario; (c) suitability map generated by MaxEnt algorithm under

future climatic scenarios and **(d)** suitability map generated by the GARP algorithm under future climatic scenarios.

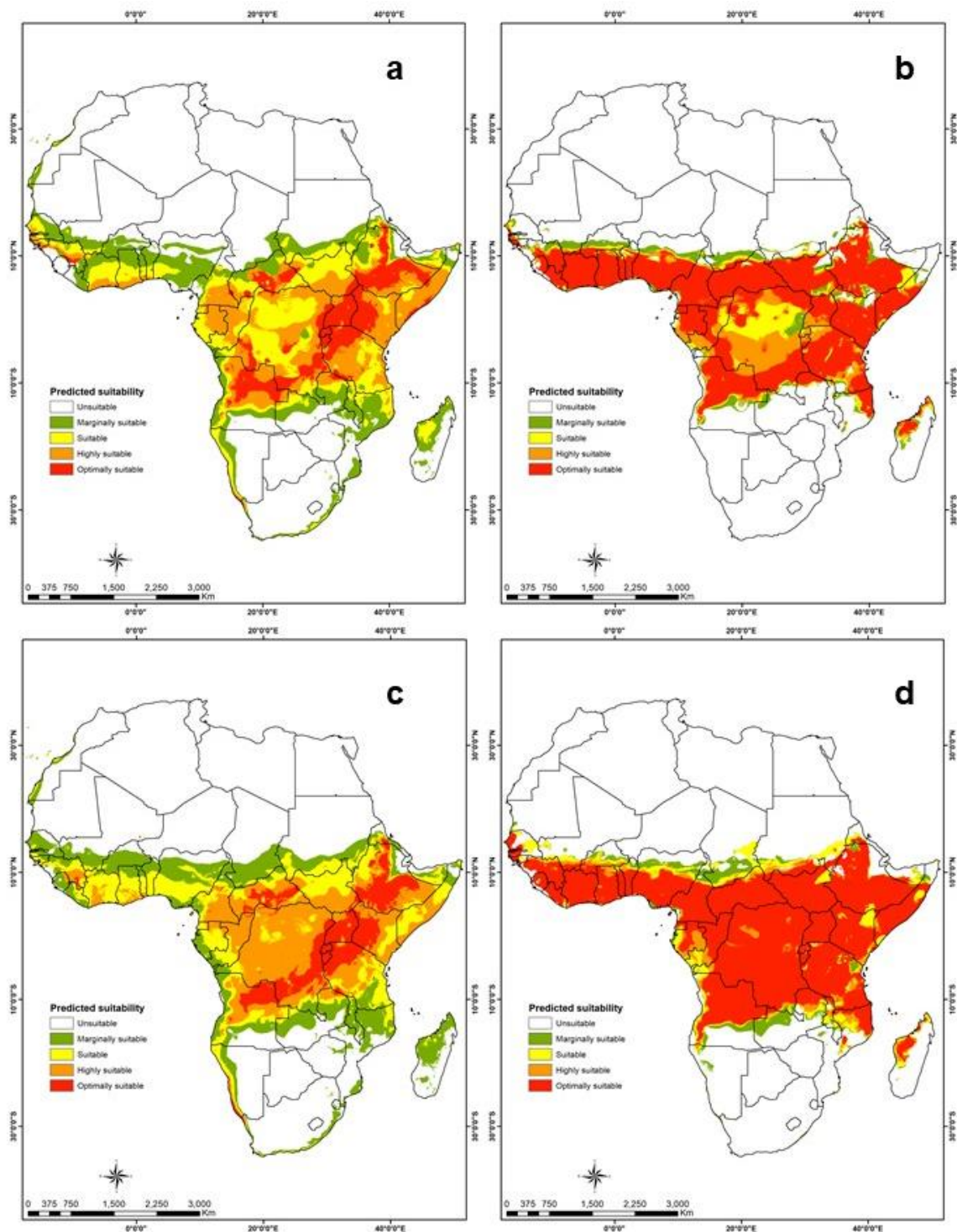


Figure 15. Predictive suitability map of the geographic distribution of *Cirina forda*. **(a)** Suitability map generated by the MaxEnt algorithm under current climate scenario; **(b)** suitability map generated by the GARP algorithm under current climatic scenario; **(c)** suitability map generated by MaxEnt algorithm under future

climatic scenarios and (d) suitability map generated by the GARP algorithm under future climatic scenarios.

4.4.6 Host Plants of Edible Saturniids in Kenya

The caterpillars were observed to show very distinct host plant specificity across the various sites surveyed. We observed that *Go. cocaulti* and *B. alcinoe* mainly feed on *Vachellia nilotica* (L.) P.J.H. Hurter and Mabb (Fabaceae), while *B. alcinoe* was feeding on *Balanites aegyptiaca* Linn (Zygophyllaceae) and *Balanites glabra* Mildbr. and Schltr. (Zygophyllaceae). *Gonimbrasia cocaulti* also fed on *Vachellia tortilis* (Forssk.) Galasso and Banfi (Fabaceae), while *Go. zambesina* and *Go. belina* on *Anacardium occidentale* Lin (Anacardiaceae) and *Go. zambesina* as well on *Mangifera indica* L. (Anacardiaceae). *Gonimbrasia krucki* was only observed feeding on *Schinus terebinthifolia* Raddi and *Schinus molle* L. (Anacardiaceae), while *C. forda* seemed to have a broader host range with *Euclea divinorum*, *Acacia mearnsii* De Wild (Fabaceae) and *Manilkara sulcata* Engl. (Sapotaceae) (Table 9).

Table 9. Host plants consumed by edible saturniids in Kenya.

Saturniid	Host Plant
<i>Gonimbrasia zambesina</i>	<i>Mangifera indica</i> , <i>Anacardium occidentale</i>
<i>Cirina forda</i>	<i>Euclea divinorum</i> , <i>Acacia mearnsii</i> , <i>Manilkara sulcata</i>
<i>Gonimbrasia cocaulti</i>	<i>Vachellia tortilis</i> , <i>Vachellia nilotica</i>
<i>Bunaea alcinoe</i>	<i>Balanites aegyptiaca</i> , <i>Balanites glabra</i>
<i>Gonimbrasia krucki</i>	<i>Schinus terebinthifolia</i> , <i>Schinus molle</i>
<i>Gonimbrasia belina</i>	<i>Anacardium occidentale</i>

In terms of genetic analyses, *E. divinorum*, *A. occidentale*, *M. indica*, *Balanites* sp., *V. tortilis*, *Manilkara* sp., *A. mearnsii*, *S. terebinthifolia* and *V. nilotica* had a 97.06–100% similarity to GenBank sequences (Table 10).

Table 10. Identities of edible saturniid host plants based on similarities with sequences from GenBank.

Collection Site	Sample Code	Species	% Similarity to GenBank Sequences	ID of Similar Sequences
Gilgil, Nakuru	HP7	<i>Euclea divinorum</i>	100	DQ924074.1
Mbita, Homabay	HP17	<i>Euclea divinorum</i>	97.06	DQ924074.1
Mbita, Homabay	HP18	<i>Euclea divinorum</i>	98.11	DQ924074.1
Gilgil, Nakuru	HP37	<i>Euclea divinorum</i>	99.64	DQ924074.1
Embu	HP38	<i>Euclea divinorum</i>	99.76	DQ924074.1
Gilgil, Nakuru	HP39	<i>Euclea divinorum</i>	100	DQ924074.1
Embu	HP40	<i>Euclea divinorum</i>	99.76	DQ924074.1
Matuu, Machakos	HP21	<i>Vachellia tortilis</i>	99.54	AF274140.1
Taita	HP22	<i>Vachellia tortilis</i>	99.77	AF274140.1
Makueni	HP23	<i>Vachellia tortilis</i>	99.77	AF274140.1
Mwingi	HP24	<i>Vachellia tortilis</i>	99.88	AF274140.1
Ngong, Kajiado	HP25	<i>Acacia mearnsii</i>	99.88	HM020723.1
Ngong, Kajiado	HP26	<i>Acacia mearnsii</i>	99.76	HM020723.1
Ngong, Kajiado	HP27	<i>Acacia mearnsii</i>	100	HM020723.1
Ngong, Kajiado	HP28	<i>Acacia mearnsii</i>	100	HM020723.1
Kilifi, Malindi	HP16	<i>Manilkara</i> sp.	99.40	DQ924092.1
Kilifi, Malindi	HP33	<i>Manilkara</i> sp.	99.40	DQ924092.1
Kilifi, Malindi	HP36	<i>Manilkara</i> sp.	99.40	DQ924092.1

Muhaka, Kwale	HP11	<i>Anacardium occidentale</i>	100	KY635877.1
Malindi, Kilifi	HP10	<i>Mangifera indica</i>	100	KX871231.1
Mbita, Homabay	HP3	<i>Balanites</i> sp.	99.35	JX517722.1
Mbita, Homabay	HP32	<i>Balanites</i> sp.	99.22	JX517722.1
Matuu, Machakos	HP45	<i>Balanites</i> sp.	99.48	JX517722.1
Matuu, Machakos	HP46	<i>Balanites</i> sp.	99.48	JX517722.1
Nanyuki	HP47	<i>Balanites</i> sp.	99.48	JX517722.1
Embu	HP48	<i>Balanites</i> sp.	99.48	JX517722.1
Embu	HP45	<i>Balanites</i> sp.	99.48	JX517722.1
Matuu, Machakos	HP2	<i>Vachelia nilotica</i>	99.30	KY10024.1
Buruburu, Nairobi	HP49	<i>Schinus terebinthifolia</i>	100	KP149521.1
Buruburu, Nairobi	HP51	<i>Schinus terebinthifolia</i>	100	KP149521.1
Kasarani, Nairobi	HP63	<i>Schinus terebinthifolia</i>	100	KP149521.1
Kasarani, Nairobi	HP62	<i>Schinus terebinthifolia</i>	100	KP149521.1
Kasarani, Nairobi	HP60	<i>Schinus terebinthifolia</i>	99.53	KP149521.1
Kasarani, Nairobi	HP61	<i>Schinus terebinthifolia</i>	99.53	KP149521.1
Kasarani, Nairobi	HP58	<i>Schinus terebinthifolia</i>	99.41	KP149521.1

4.5 Discussion

This study has documented seven species of edible saturniids in Kenya. Three saturniid species, i.e., *C. forda*, *Go. zambesina* and *B. alcinoe*, are consumed in Kenya, mainly along the coastal belt along the Giriama community. Moreover, *C. forda* is widely consumed in West, Central and southern Africa (Ande 2002; Badanaro et al. 2014; Dwomoh et al. 2010; Mabossy-Mobouna et al. 2016). *Bunaea alcinoe* is also a popular edible insect in West and Central Africa, for instance in countries like DR Congo, Cameroon and Nigeria (Amadi et al. 2005; Mbata et al. 2002; Temitope et al. 2014), while *Go. zambesina* is highly popular in southern and Central Africa (Kachapulula et al. 2018; Latham 2015; Mbata et al. 2002). *Gonimbrasia krucki* is widely consumed in DR Congo (Latham 2015), but not in Kenya. For *Go. cocaulti*, no records of human consumption are available from Kenya or elsewhere in Africa. However, due to the similarity of *Go. cocaulti* larva to that *Go. belina*, it is likely misidentified, given that they have been observed in consignments of mopane caterpillar in the UK (Siozios et al. 2020).

Bunaea alcinoe, though black in colour and commonly found in Kenya, its genetic configuration is different from the red forms collected in Nigeria (Akanbi 1973). In Nigeria and DR Congo, both color forms have been reported feeding on the same host plant (Akanbi 1973; Latham 2015), yet this is the first time their genetic difference has been assessed. Further detailed studies relating the morphological and molecular differences, mating compatibility between color forms of *B. alcinoe* can shed light on their taxonomic status.

The sampled *G. zambesina* moths also exhibited two color forms and juveniles of the green adults carried black spines, as previously reported (Pinhey 1956) and in <http://www.africanmoths.com/> (accessed on 26 September 2019) (Goff 2019). The brown moths emerged from red spined larvae, which had been previously described as *G. said* (Pinhey 1956), but inconclusively. Identification of edible saturniid species is important for the purpose of conservation (Thomas 2013; Vantomme et al. 2004), maintenance of quality in production (Siozios et al. 2020) as well as mainstreaming consumption of the caterpillars.

We observed a bivoltine lifecycle in *C. forda*, with larvae occurring in April–June and October–December. In contrast, *C. forda* has been recorded as univoltine in Togo and Nigeria with larval occurrence between July and September (Ande and Fasoranti

1997; Badanaro et al. 2014) and in DR Congo with larvae appearing between November and January (Latham 2015). In all these cases, the occurrence of *C. forda* larvae coincides with the rainy seasons. For, *B. alcinoe* we noted a bivoltine lifecycle with larval appearance between April–May and October–December. In contrast, the same species in DR Congo is univoltine and occurs between October and May (Latham 2015). Understanding the temporal distribution of edible saturniids informs the need for mass production to ensure a continuous source throughout the year.

The model for habitat suitability of *B. alcinoe* and *C. forda* demonstrates that the two species thrive well within the tropical regions of Africa. However, *B. alcinoe* spreads slightly into the subtropics, specifically in southern Africa. The model concurs with previous reports of availability and consumption of the two edible saturniids in southern, central and western Africa (Badanaro et al. 2014; Dithlogo 1996; E. A. Dwomoh et al. 2010; Kelemu et al. 2015; Kwiri et al. 2020; Latham 2015; Mabossy-Mobouna et al. 2016; Mbata et al. 2002). However, the availability of *C. forda* in the southern African region is not concurrent with previous reports (Oberprieler 1995; Pinhey 1975) which recorded a wide distribution in the southern Africa region. This could be due to the limited data on the presence of the two saturniid species in the GBIF database which was used in this study. Future predictions for both species show a reduction in habitat suitability, stressing the need to conserve saturniid habitats.

The saturniids identified fed on specific host plants and consequently their availability depends on the occurrence of these host plants. We found *B. alcinoe* feeding on *B. aegyptiaca* and *B. glabra* similar to reports in Nigeria (Akanbi 1973). However, other research suggests a wide range of host plants, e.g., for DR Congo with *Sarcocephalus latifolius* (JE Sm.) EA Bruce (Rubiaceae), *Acacia auriculiformis* A. Cunn. ex Benth. (Fabaceae), *Dacryodes edulis* (G. Don) H.J.Lam. (Burseraceae), *Crossopteryx febrifuga* (Afzel. ex G.Don) Benth. (Rubiaceae) and *Anthocleista schweinfurthii* Gilg (Loganiaceae) (Pinhey 1972), and in Nigeria on *Holarrhena floribunda* (G. Don) Durand and Schinz (Apocynaceae), *Ekebergia sengalensis* A Juss. (Meliaceae), *Fragraea fragrans* Roxb. (Gentianaceae), *Cleistopholis patens* (Benth.) Engl. and Diels (Annonaceae) and *Spondias mombin* L. (Anacardiaceae) (Akanbi 1973). In our study in Kenya we observed, for the first time, larvae of *C. forda* feeding on *E. divinorum*, *A. mearnsii* and *Manilkara sulcata*, while in DR Congo it feeds mainly on *Crossopteryx febrifuga* (Latham 2015). In West Africa, *C. forda* is confined on the shea butter tree, *Vittelaria paradoxa* (Ande and Fasoranti 1997; Badanaro et al. 2014; E. A.

Dwomoh et al. 2010), whereas in southern Africa host plants include *Burkea africana* Hook (Fabaceae) and *Albizia versicolor* Welw. ex Oliv. (Fabaceae) (Oberprieler 1995). We collected *Go. zambesina* from mango and cashew nut trees in Kenya, corroborating earlier findings (Pinhey 1956). Cashew nut and mango trees are important commercial trees whose nuts and fruits, respectively, are widely consumed. *Gonimbrasia zambesina* is sometimes considered a pest of mango trees (de Almeida 1972). Spraying of mango trees to curb pests may pose a threat to *Go. zambesina* larvae feeding on the leaves.

Apart from being host plants for edible insects, most of these plants have other uses in communities in Kenya and beyond. For example, *E. divinorum* is utilized by the Maasai in Kenya as firewood, their stem cuttings are used as toothbrushes and their fruits are edible (Bussmann et al. 2006). Marakwets from the Rift Valley region in Kenya use *E. divinorum* as anti-venom (Kipkore et al. 2014) while the Luo from western Kenya use it to treat venereal diseases (Johns et al. 1990). Maasai also use *V. tortilis* and *V. nilotica* for firewood (Tian 2017) while the Marakwet employ them for treating abdominal pains (Kipkore et al. 2014). *Balanites* spp. are used to treat coughs (Johns et al. 1990), and finally *A. mearnsii* is often planted for firewood, timber, apiculture and a source of tanning dyes, and trees are also used for shade, nitrogen fixation and controlling soil erosion (Orwa et al. 2009). Such traditional knowledge can be used to encourage communities to conserve these plants and hence protect habitats for edible saturniid species.

4.6 Conclusions

We successfully documented seven species of saturniids in Kenya, among which three are consumed. The identity of these species was confirmed both at molecular and morphological level. Their distribution, seasonality and host plants were also established. We emphasize the importance of combining molecular barcoding, morphological identification, phenology, and ecology studies in identification of edible saturniid species. Potential habitats under current and future climate scenarios of two edible saturniid species, *B. alcinoe* and *C. forda*, were mapped. This information may help in implementing conservation measures for edible saturniids and their host plants. Due to their seasonal occurrence, further research is required on prospects for mass production to ensure a continuous supply and to prevent overharvesting from the wild forest for enhance sustainability. Moreover, potential economic benefits of edible

saturniids for local communities in East Africa need to be quantified and their value chains established.

5.0 Parasitoids and entomopathogens of edible saturniids and the potential for mass rearing of selected saturniid species in Kenya

5.1 Abstract

Edible saturniids that belong to the order *Lepidoptera* constitutes 60% of edible insects consumed in Africa. They are highly nutritious and a source of income supporting livelihoods in Africa. Despite this available information on their natural regulatory factors such as parasitoids and entomopathogens in Kenya that control their populations in the wild is scanty, which this study aimed to address. This is critical to develop mass rearing of edible saturniids. Samples of saturniid larvae and eggs collected from Homabay, Nairobi, Machakos, Makueni, Nakuru and Kitui between December 2016 and September 2018 were reared in the laboratory at *icipe*. Parasitoids that emerged from eggs, larvae and pupae were collected and pinned for identification. Dead larvae collected in the field or those that died in the laboratory were examined for bacterial and fungal entomopathogens. The bacterial and fungal entomopathogens were isolated on Nutrient/Mackonkey agar and Potato dextrose agar (PDA), respectively. Pure cultures of the entomopathogens were characterized using molecular tools.

Two edible saturniid species were selected for this study namely: *Go. zambesina* and *C. forda*. Larvae were collected in the wild and reared in cages while feeding on their respective host plants until they developed into pupae then adult moths. Eggs laid by the moths were used to set up life cycle experiments.

Parasitoids observed in this study belong to the orders Diptera and Hymenoptera. Dipteran larval pupal parasitoids included, tachinids, *Senometopia* sp. (cf. *evolans* Wiedemann) that emerged from *Gonimbrasia zambesina*, *Gonimbrasia krucki* and *Gonimbrasia gueinzii*; a new *Ceromyia* sp. (Robineau-Desvoidy) from *Bunaea alcinoe*, *Gonimbrasia belina* and *Gynasia maja*; an unidentified tachinid parasitoid from *Cirina forda* and a *Sarcophaga* sp. from *B. alcinoe*. A large Ichneumonid parasitoid *Euryophion pisinnus* (Gauld & Mitchell) was also frequently encountered in *B. alcinoe*. Larval parasitoid constituted braconids including a *Cotesia* sp. from *G. zambesina*, *G. belina* and *B. alcinoe*; *Aleiodes trifasciatus* (Enderlein) from *G. zambesina*; *Glyptapanteles maculitarsis* from *B. alcinoe* and a *Microgastrinae* sp. from *C. forda*.

Egg parasitoids belonging to the sub family Entedoninae of Eulophidae and *Eupelmus* sp. (Eupelmidae) were observed on *B. alcinoe* and *G. krucki*, respectively.

Thirteen bacteria species were isolated among which nine were potential entomopathogens. They include *Enterococcus mundtii*, *Bacillus cereus*, *Staphylococcus sciuri*, *Staphylococcus gallinarum*, *Pseudomonas putida*, *Enterobacter hormaechei*, *Enterococcus faecalis*, *Alcaligenes faecalis* and *Stenotrophomonas maltophilia*. The isolates were 97-99% similar to sequences already submitted to the GenBank.

The complete life cycle of *G. zambesina* was 152-170 days, while that of *C. forda* ranged from 102-246 days. Male moth emerged earlier than female moths for both species. These findings will inform the prospects of rearing edible saturniids both for research and consumption.

Key words: Saturniidae, Tachnidae, Ichneumonidae, Braconidae, entomopathogenic bacteria

5.2 Introduction

Edible saturniids form a large part of caterpillars consumed in Africa (Kelemu et al. 2015). They are highly nutritious providing vital vitamins and proteins to households, especially women and children. Nutritionally, the saturniids are rich in proteins which often make up more than 50% of their body weight (Omotoso 2006). Edible saturniids are also a major source of income in rural sub-Saharan Africa (Balinga et al. 2004; Glew et al. 1999; Makhado et al. 2014; Omotoso 2006; Osasona and Olaofe 2010). A good example is the commercialization of *Gonimbrasia belina* with about 63% of the caterpillars harvested being sold in Limpopo, South Africa (Baiyegunhi and Oppong 2016). The most commonly consumed saturniid caterpillars include *B. alcinoe*, *C. forda*, *G. belina* and *Cirina butyrospermi* among others (Adepoju and Daboh 2013; Baiyegunhi and Oppong 2016; Kelemu et al. 2015; Latham 2015). The United Nations has termed edible insects as a solution to curb food insecurity (FAO 2013). *Gonimbrasia belina* has been recommended as a potential source of protein in fortified blended foods. This is due to its crude protein level of about 55% compared to other protein sources like common beans, soybeans, cow peas and groundnuts (Kwiri et al. 2014).

Saturniid caterpillars are seasonal. They occur once or twice a year depending on the region; usually following the rainy season. For instance, *C. forda* is univoltine in Togo and Nigeria where the larvae appear between July and September (Ande and Fasoranti 1997; Badanaro et al. 2014) while in Congo they appear between November and January (Latham 2015). On the other hand, *G. belina* is mostly bivoltine in Botswana (Ditlhogo 1996). The seasonality of edible saturniid caterpillars and dependence on wild harvests, which is increasingly becoming unreliable is a major setback in the effort to mainstream their consumption. Hence the need to develop methods for mass production of edible saturniids. Understanding natural regulatory factors of saturniids such as parasitoids and entomopathogens, that could also constrain their mass rearing is critical.

Edible saturniids are also host-specific, hence their availability is dependent on the availability of particular host plants. Their host plants are mainly shrubs, forest trees and domesticated fruit trees (Latham 2015; Mabossy-Mobouna et al. 2016; Thomas 2013). For example, the mopane worm (*Gonimbrasia belina*) feeds mainly on the mopane tree *Colophospermum mopane* (Kirk ex Benth.) Kirk ex J.Léonard (Fabaceae) in southern Africa (Baiyegunhi and Oppong 2016; Thomas 2013). On the other hand, the shea defoliator (*Cirina forda*) feeds exclusively on the shea tree (*Vitellaria paradoxa*) in West Africa (Agbidye and Nongo 2012; Odebiyi et al. 2011).

Over time, commercialization of edible saturniid caterpillars has led to overharvesting and destruction of habitats (Ngute et al. 2020). A good example is *C. forda* host plants *Erythropheum suaveolens* and *Baillonella toxiperma* Pierre (Sapotaceae) which are also good candidates for timber. Their logging destroys the natural habitat of *C. forda*. Breaking branches during harvesting is also common among edible saturniid caterpillar harvesters. They also overharvest the caterpillars which reduces future generations. All these changes point to a need for mass rearing of edible saturniid caterpillars.

There have been efforts to rear edible saturniids in captivity. However, there has been challenges like pupal diapause (Bama et al. 2018) which also occurs in the wild. This study aims to understand life cycles of two edible saturniid caterpillar species namely *C. forda* and *G. zambesina*. Edible saturniids face several natural regulatory factors including parasitoids, pathogens, predators and human intervention. About 350 species of parasitoids have been recorded in 175 species of Saturniidae. They include 21 Braconidae, 115 Tachinidae, 2 Pyralidae, 105 Ichneumonidae, 4 Sarcophagidae

and about 100 from the Chalcidoidea and Proctotrupoidea families (Peigler 1994). Hymenopteran chalcid parasitoid wasps, *Hockeria crassa*, *Megaselia scalaris* (Dwomoh et al. 2004) and *Hockeria* spp. (Walker) (Muhammad and Ande 2014) parasitize the pupa of *C. forda* (Dwomoh et al. 2004). *Hockeria* spp. (Walker) also parasitizes the pupae of *G. belina*, while *Hockeria crassa* parasitizes the pupa of *Imbrasia macrops* (Rebel). The two also attack the pupa of *Gonimbrasia cytherea cytherea* (Boucek 1974). *Imbrasia epimethea* is parasitized by the Tachinidae *Ceromya luteicornis*. *B. alcinoe* on the other hand is parasitized by *Carcelia* sp. (Tachinidae), *Eucepsis* sp. (Chalcididae), *Sarcophaga* sp. (Sarcophagidae) and *Ceromya luteicornis* (Tachinidae) (Akanbi 1973).

A Braconid, *Glyptapanteles maculitarsis* has been observed emerging from the larvae of *Imbrasia cytherea*, *Imbrasia zambesina*, *Imbrasia tyrreha*, *B. alcinoe* and *Gy. maja* (Geertsema 1975; Van Den Berg 1974; Walker et al. 1990). Chalcid, Tachinid and Ichneumonid parasites attacking *G. belina* larvae have also been reported in Botswana (Ditlhogo 1996).

Studies carried out in Nigeria showed that the eggs of *C. forda* were destroyed by a parasitoid, *Anastatus* spp. that belongs to the Chalcididae (Muhammad and Ande 2014) and *Entedon* sp. belonging to Eulophiidae (Odebiyi et al. 2011). Four egg parasitoids namely *Mesocomys pulchriceps* Cam., *Eupelmus urozonus* (Dal.), *Anastatus* spp. (Eupelmidae), and *Pediobius* spp. (Eulophidae) were recorded for *G. belina* in the northern Transvaal region in South Africa (Van Den Berg 1974; 1974). Several fungal species were also found to attack *Cirina forda* in Nigeria. They include *Nomuraea rileyi* (Hypocreales, family Clavicipitaceae), *Beauveria bassiana* (Hypocreales: Cordycipitaceae) (Muhammad and Ande 2014), *Aspergillus niger*, *Aspergillus flavus*, *Trichoderma* sp. and *Fusarium solani* (Odebiyi et al. 2011). Two viruses, *Nucleopolyhedrovirus* (NPV) and *Granulovirus* (GV) also infect the larvae of *Cirina forda* (Muhammad and Ande 2014).

Despite availability of edible saturniids in Kenya (Kioko et al. 2007), information on parasitoids and entomopathogens attacking them is scanty. This study sought to document parasitoids and entomopathogens of edible saturniids in Kenya.

5.3 Materials and Methods

5.3.1 Study area and sample collection

This study was carried out in Kenya between December 2016 and September 2018. Samples of Saturniid caterpillar were collected from Homabay, Nairobi, Machakos, Makueni, Nakuru and Kitui. Insect larvae and eggs were collected from the field and transported to the laboratory at *icipe*.

5.3.2 Parasitoid collection and identification

Saturniid larvae were kept in cages and fed on their respective host plant leaves. Fresh leaves were introduced daily. Larvae were observed for emergence of larval parasitoids. The parasitoids that emerged were collected, processed and pinned for identification. Other larvae were fed until pupation, when trays containing sterile sawdust were offered as pupation medium. The pupae were collected and placed on a tray lined with paper towel and occasionally sprinkled with water to keep the environment moist. The pupae were placed in cages and observed for emergence of parasitoids or saturniid moths. Pupae collected from the field were treated in the same manner. Once the parasitoids emerged, they were collected, processed and pinned for identification at the Biosystematics unit in *icipe*. Eggs collected in the wild were also kept in plastic containers and observed for parasitoid emergence. Egg parasitoids were also pinned for identification at the Biosystematics unit in *icipe*. Identification of tachniids was done by Prof. Pierfilippo Cerretti while the other parasitoids were identified by Dr. Robert Copeland. Parasitism studies were not carried out since we lacked stable insect colonies in the laboratory.

Dead insect larvae collected both in the field and the rearing facility were subjected to isolation of possible entomopathogens.

5.3.3 Isolation of entomopathogens

Fungi

Potato dextrose agar (PDA) was used for isolation of fungi from the dead larvae. PDA was prepared, autoclaved and dispensed in petri dishes. An antibiotic (Streptomycin) was added to the media to prevent bacterial growth. A small piece of the larvae was cut, surface sterilized and placed on PDA under sterile conditions and incubated at 25°C. The plates were observed daily for growth. To surface sterilize, the larva was dipped in 1.5% bleach followed by 70% ethanol after which it was rinsed three times

in distilled water. The insect was then blotted on filter paper to dry the water. Different fungi grew on the plate and were isolated to obtain pure cultures. Fresh PDA was prepared, autoclaved and dispensed in petridishes. A scalpel sterilized on a flame was used to cut a small piece in the middle of each fungi colony and plated on a new separate petridish. The petridishes were sealed with parafilm and incubated at 25°C to get pure cultures. Once grown, the fungi were stored at 4°C to arrest growth awaiting molecular identification.

Bacteria

To isolate bacterial entomopathogens, dead larvae were surface sterilized before plating on nutrient agar. To surface sterilize, the larva was dipped in 1.5% bleach followed by 70% ethanol after which it was rinsed three times in distilled water. The insect was then blotted on filter paper to dry the water. A piece of the dead larva was cut near the abdomen and placed on the agar. The plates were incubated for 24 hours at 37°C. The different colonies were isolated on nutrient agar and MacConkey agar. Each pure colony was multiplied in LB broth. Upon inoculation with the bacteria, the LB broth was rocked for 24 hours. The bacteria were spun at 8000rpm at 4°C to obtain a pellet. The pellet was stored at -80°C awaiting molecular characterization.

Pathogenicity studies were not carried out due to lack of a stable insect colony in the laboratory.

5.3.4 Molecular identification of entomopathogens

Tissue preparation, DNA extraction and quantification

The fungus sample was scrapped off the media under sterile conditions. About 0.2g of fungus sample was placed in a 2ml tube containing ceramic beads, lysis buffer PA1 (Bioline, UK) and RNase A and crushed for 3 minutes in a Tissue lyser II (Qiagen USA). Fungal genomic DNA was extracted using Isolate II Plant DNA extraction Kit (Bioline, UK) according to manufacturer's instructions.

The bacteria pellet attained after spinning down the cells in LB broth was placed in a 2ml reaction tube. A portion of the insect parasitoid sample was cut using a sterile blade and placed in a 2ml tube. Genomic DNA for bacteria and insect parasitoids was extracted using Isolate II genomic DNA extraction kit (Bioline, UK) according to the manufacturer's protocol. The resultant DNA from bacteria and fungi was eluted in 50µl Elution buffer (Bioline, UK) and quantified using a NanoDrop 2000/2000c

Spectrophotometer (Thermo Fisher Scientific, Wilmington, USA). Bacteria and fungi DNA samples were stored at -20°C awaiting Polymerase Chain Reaction (PCR).

PCR for bacteria samples

PCR was conducted using 16S 27F-1492R primers (27F- 5' AGAGTTTGATCMTGGCTCAG 3'; 1492R 5' TACCTTGTTACGACTT 3'). These primers amplify a 1450 base pair fragment of the 16S rRNA gene. Isolated insect DNA was amplified in 30µl PCR mix containing 16.95µl PCR water, 6µl My Taq Buffer (Bioline, UK), (5mM dNTPs, 15mM MgCl₂, stabilizers and enhancers), 1.5µl of each primer, 0.75µl of 25mM Mgcl₂ (Thermoscientific, USA), 0.3µl 1 unit My Taq DNA polymerase (Bioline, UK) and 15ng/l of DNA template.

The reaction was set up in a Mastercycler Nexus Gradient thermocycler (Thermo scientific, USA) using conditions as follows: Initial denaturation at 95°C for 2 minutes followed by 40 cycles of denaturation at 95°C for 30 seconds, annealing at 51.9°C for 45 seconds and primer elongation at 72°C for 1 minute. The final extension step lasted for 10 minutes at 72°C.

PCR for fungi samples

For fungi identification, PCR was conducted using DNA barcoding ITS4/ITS5 primers (ITS4- 5' GGAAGTAAAAGTCGTAACAAGG 3'; ITS5- 5' TCCTCCGCTTATTGATATGC 3'). These primers amplify a 600 base pair fragment of the internal transcribed spacer region. Isolated insect DNA was amplified in 30µl PCR mix containing 16.95µl PCR water, 6µl My Taq Buffer (Bioline, UK), (5mM dNTPs, 15mM MgCl₂, stabilizers and enhancers), 1.5µl of each primer, 0.75µl of 25mM Mgcl₂ (Thermoscientific, USA), 0.3µl 1 unit My Taq DNA polymerase (Bioline, UK) and 15ng/l of DNA template.

The reaction was set up in a Mastercycler Nexus Gradient thermocycler (Thermo scientific, USA) using conditions as follows: Initial denaturation at 95°C for 1 minute followed by 35 cycles of denaturation at 95°C for 30 seconds, annealing at 59°C for 40 seconds and primer elongation at 72°C for 1 minute. The final extension step lasted for 10 minutes at 72°C.

Agarose Gel electrophoresis, PCR product purification and sequencing

Agarose (1%) gel stained with ethidium bromide (10mg/ml) was used to resolve the PCR products. Electrophoresis followed at 80 volts for 1 hour (Bio-Rad model 200/2-0 power supply, Bio-Rad laboratories Inc., USA). DNA bands were visualized using an ultraviolet transilluminator and photographed using the KETA GL imaging system software (Wealtec Corp., USA).

The resultant PCR products for both the insect parasitoids, bacteria and the fungi were purified using QIAquick PCR purification kit (Qiagen, Germany) and quantified with a NanoDrop 2000/2000c Spectrophotometer (Thermo Fisher Scientific, Wilmington, USA). They were sent for bidirectional sequencing at Macrogen Inc. (Netherlands).

Sequence analysis

Bacteria, fungi and insect sequences were assembled and edited using Bioedit software v 7.0.5.2 (Hall 1999). A consensus sequence generated from both the forward and reverse strand was queried on Basic Local Alignment Search Tool (BLAST) (Altschul et al. 1990) to determine similarity with already submitted sequences. Multiple sequence alignments were created on Clustal W (Thompson et al. 1994). Phylogenetics and molecular evolutionary analysis was done using Mega X (Kumar et al. 2018). The evolutionary history was inferred by using the Maximum Likelihood method and Tamura-Nei model (Tamura and Nei 1993).

5.3.5 Insects for life cycle studies

Gonimbrasia zambesina and *Cirina forda* larvae (3rd and 4th instar) were collected from the field and transported to the International Centre of Insect Physiology and Ecology (*icipe*). *Gonimbrasia zambesina* was maintained at the *icipe* station in Nairobi, while *Cirina forda* were maintained at the *icipe* station in Mbita in Western Kenya.

The larvae were reared in Perspex cages (50x50x50cm) netted on the sides to improve ventilation. The cages had metallic stands that were dipped in containers filled with tap water to prevent crawling insects from getting into the cage. The larvae were reared at 25°C and exposed to a 12h:12h photoperiod. Twigs of the host plants were arranged in a bouquet, fastened with cottonwool, and dipped in water to maintain moisture (Figure 16). The twigs were changed daily, and droppings cleaned up to keep the cage environment fresh.

The larvae fed until they reached the prepupal stage. At this stage, they stopped feeding, reduced movement, and settled at the bottom of the cage. At this stage, they were transferred to plastic trays containing moist sawdust to burrow and pupate. The sawdust was kept moist by sprinkling water daily. These trays with the pupae were placed in Perspex cages (50x50x50cm) awaiting emergence of adult moths. The large cages were necessary to allow the moths to spread out their wings freely after emergence. The moths emerged, mated and laid eggs. The eggs laid were used for life cycle experiments.



Figure 16. *Gonimbrasia zambesina* larvae feeding on mango tree leaves in a cage set-up mimicking a tree in the wild.

5.3.6 Life cycle experiments

The eggs laid in each day was recorded and monitored until larvae emerged. Once the larvae emerged, they were transferred into one litre plastic containers. One larva was placed in each container separately. Leaves of the host plant were placed in each container for the larva to feed (Figure 17). Sixty insects were placed in different jars for each life cycle generation. The leaves were replaced, and the droppings cleaned daily. Data was recorded every morning. Upon reaching the prepupa stage, the insects were transferred to trays containing sawdust where they burrowed and pupated. The sawdust was sprinkled with water to keep it moist. The pupae were then sexed by observing the pattern at the rear end of the pupae. The trays were placed in cages to

offer enough room for the moths to emerge. The dates for pupation and adult moth emergence were recorded. The sex of the moths was determined by hairy antennae for males and lack thereof for females.



Figure 17. *Gonimbrasia zambesina* larvae feeding on mango tree leaves in separate jars in a lifecycle experiment.

5.4 Results

5.4.1 Parasitoids

Larval-pupal parasitoids

Larval-pupal parasitic dipterans belonging to the Tachinidae such as *Senometopia* sp. (cf. *evolans* Wiedemann) emerged from *G. zambesina* and *G. krucki*. An undescribed species of *Ceromyia* sp. (Robineau-Desvoidy) from *B. alcinoe*, *G. belina* and *G. maja* was recorded. Another undescribed species of Tachinid parasitoid emerged from *C. forda* pupae. Large Ichneumonid larval parasitoid, *Euryophion pisinnus* Gauld & Mitchell and a Dipteran *Sarcophaga* sp. were frequently encountered from *B. alcinoe* (Table 11).

Larval parasitoids

Braconids belonging to *Cotesia* sp. were observed on *G. zambesina*, *G. belina*, and *B. alcinoe*. Braconid larval parasitoids, *Aleiodes trifasciatus* (Enderlein) was observed to emerge from 2nd and 3rd larval instars of *G. zambesina*. Other braconids observed

included *Glyptapanteles maculitarsis* (Figure 18) and an undescribed species belonging to subfamily Microgastrinae on *B. alcinoe* and *C. forda* larvae, respectively. (Table 11).

Egg parasitoids

Egg parasitoids belonging to Eupelmidae and an Eulophid parasitoid belonging to the subfamily Entedoninae were observed for the first time on *G. krucki* and *B. alcinoe*, respectively (Table 11).

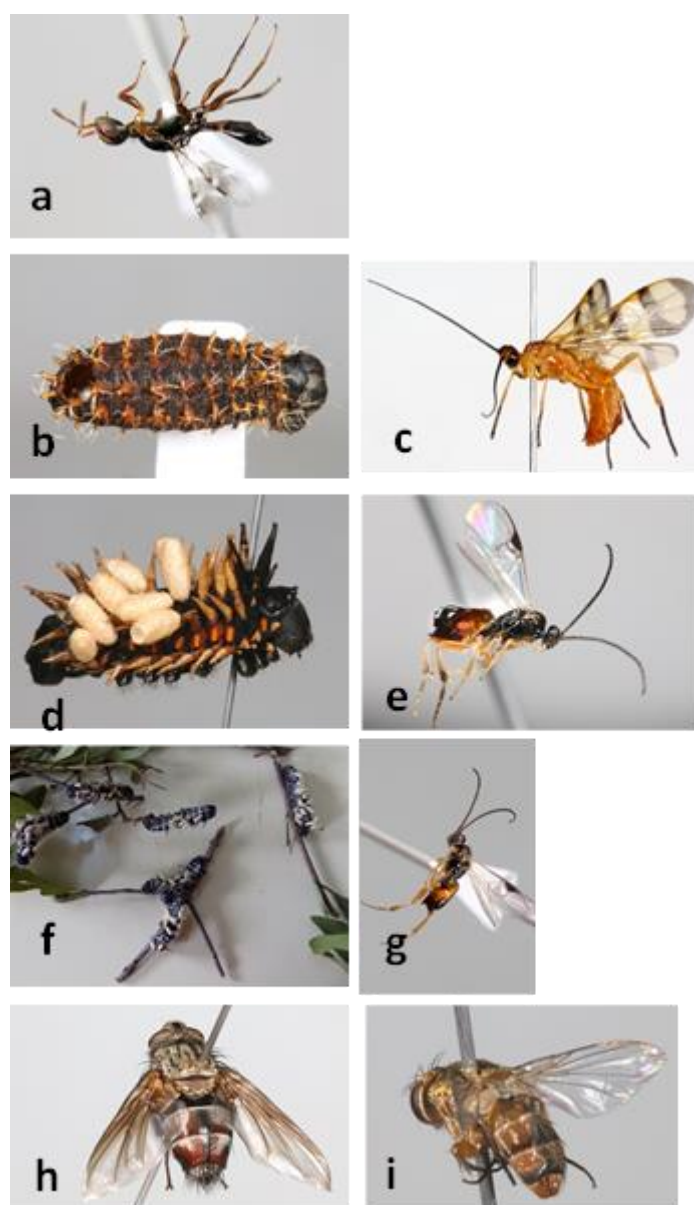


Figure 18. Parasitoids of saturniids. **a)** Undescribed egg parasitoid of *Gonimbrasia krucki*, belonging to Eupelmidae. **b)** *Gonimbrasia zambesina* mummy, parasitized with *Aleiodes fasciatus*. **c)** *Aleiodes fasciatus*, a larval-pupal parasitoid of *Gonimbrasia zambesina*. **d)** *Bunaea alcinoe* larvae parasitized by *Glyptapanteles maculitarsis* **e)** *Glyptapanteles maculitarsis*, a larval-larval parasitoid of *Bunaea alcinoe* **f)** *Cirina forda* larvae parasitized by an undescribed Microgastrinae **g)** Undescribed Microgastrinae, a larval parasitoid of *Cirina forda* **h)** Undescribed parasitoid belonging to Tachinidae that emerged from *Cirina forda* pupa **i)** *Bunaea alcinoe* tachinid parasitoid

Table 11. Parasitoids affecting different saturniid species

Order: Family	Parasitoid	Saturniid host	Life stages affected
Diptera: Tachinidae	<i>Senometopia</i> sp. (cf. <i>evolans</i> Wiedemann)	<i>Go. zambesina</i>	Larval-pupal
Diptera: Tachinidae	<i>Ceromyia</i> sp. (Robineau-Desvoidy)	<i>B. alcinoe</i> , <i>Go. belina</i> , <i>Go. maja</i>	Larval-pupal
Hymenoptera: Ichneumonidae	<i>Euryophion pisinnus</i> Gauld & Mitchell	<i>B. alcinoe</i> , <i>Go. krucki</i>	Larval-pupal
Diptera: Sarcophagidae	<i>Sarcophaga</i> sp.	<i>B. alcinoe</i>	Larval-pupal
Diptera: Tachinidae	Undescribed.	<i>C. forda</i>	Larval-pupal
Hymenoptera: Braconidae	Undescribed Micogastrinae.	<i>C. forda</i>	Larval
Hymenoptera: Braconidae	<i>Glyptapanteles maculitarsis</i> ,	<i>B. alcinoe</i>	Larval
Hymenoptera: Braconidae	<i>Aleiodes trifasciatus</i> (Enderlein)	<i>G. zambesina</i>	Larval
Hymenoptera: Braconidae	<i>Cotesia</i> sp.	<i>G. belina</i> , <i>Go. zambesina</i> , <i>B. alcinoe</i>	Larval
Hymenoptera: Eupelmidae	Undescribed species	<i>Go. krucki</i>	Egg
Hymenoptera: Eulophidae	Undescribed Entedoninae	<i>B. alcinoe</i>	Egg

5.4.2 Bacteria

Thirteen species of bacteria were isolated and identified from dead saturniid larvae. Among these, nine were potential entomopathogens according to previous published information on their pathogenicity to other insects. *Enterococcus mundtii*, *Bacillus cereus*, *Staphylococcus sciuri*, *Staphylococcus gallinarum*, *Pseudomonas putida* and *Enterobacter hormaechei* were isolated from *G. zambesina*. Entomopathogenic bacteria isolated from *B. alcinoe* included *Enterococcus faecalis*, *Staphylococcus sciuri*, *Staphylococcus gallinarum* and *Enterobacter hormaechei*. Isolates from *G. cocaulti* included *Alcaligenes faecalis*, *Stenotrophomonas maltophilia* and *Enterobacter hormaechei*, while *Staphylococcus gallinarum* was isolated from *G. krucki* (Table 12).

Enterococcus mundtii isolates were 98-99% similar to sequence AB831185.1 from the Genbank while *Enterobacter hormaechei* were 97% similar to CP031726.1. *Bacillus cereus*, *staphylococcus sciuri* and *Pseudomonas putida* showed 99% similarity to sequences MH041186.1, MK205162.1 and JN679855.1 respectively. *Staphylococcus gallinarum* was 98-99% similar to MH371285.1 while *Alcaligenes faecalis* and *Stenotrophomonas maltophilia* showed 98% similarity to MK517568.1 and MK300721.1 respectively (Table 13).

Table 12. Bacteria isolated from dead saturniid larvae

Isolated bacteria	Saturniid host	Number of isolates	Potential entomo-pathogen	Pathogenicity in other insects
<i>Enterobacter hormaechei</i>	<i>B. alcinoe</i> , <i>G. zambesina</i>	12	Yes	Leafroller weevil beetle, <i>Rhynchites bacchus</i> (Gokce et al. 2010)
<i>Bacillus amyloliquefaciens</i>	<i>G. cocaulti</i> , <i>G. krucki</i>	3	No	-
<i>Alcaligenes faecalis</i>	<i>G. cocaulti</i>	1	Yes	Gypsy moth, <i>Lymantria dispar</i> ; Lackey moth, <i>Malacosoma Neustria</i> ; House fly <i>Musca domestica</i> ; Mediterranean fruit fly, <i>Ceratitis capitata</i> (Ruiu et al. 2017)
<i>Enterococcus casseliflavus</i>	<i>G. cocaulti</i> , <i>G. krucki</i> , <i>G. zambesina</i>	5	No	-
<i>Stenotrophomonas maltophilia</i>	<i>G. cocaulti</i>	1	Yes	Gypsy moth, <i>Lymantria dispar</i> ; Lackey moth, <i>Malacosoma Neustria</i> ; House fly <i>Musca domestica</i> ; Mediterranean fruit fly, <i>Ceratitis capitata</i> (Ruiu et al. 2017)
<i>Klebsiella michiganensis</i>	<i>B. alcinoe</i>	1	No	
<i>Enterococcus mundtii</i>	<i>G. zambesina</i>	7	Yes	Gypsy moth, <i>Lymantria dispar</i> ; Lackey moth, <i>Malacosoma Neustria</i> ; House fly <i>Musca domestica</i> ; Mediterranean fruit fly, <i>Ceratitis capitata</i> (Ruiu et al. 2017)
<i>Staphylococcus sciuri</i>	<i>G. zambesina</i> , <i>B. alcinoe</i> ,	3	Yes	Mediterranean corn borer, <i>Sesamia nonagrioides</i> (ESKİ et al. 2015)
<i>Staphylococcus gallinarum</i>	<i>G. zambesina</i> , <i>G. krucki</i>	3	Yes	Silkworm, <i>Bombyx mori</i> (Han'guk-Ŭngyong-Konch'ung-Hakhoe. et al. 2003)
<i>Bacillus cereus</i>	<i>G. zambesina</i>	1	Yes	White grub, <i>Anomala dimidiata</i> (Selvakumar et al. 2007)

<i>E. coli</i>	<i>G. krucki</i>	3	No	
<i>Pseudomonas putida</i>	<i>G. zambesina</i>	1	Yes	Colorado Potato Beetle, <i>Leptinotarsa decemlineata</i> (Muratoğlu et al. 2011)
<i>Enterococcus faecalis</i>	<i>B. alcinoe</i>	1	Yes	Beet Armyworm, <i>Spodoptera exigua</i> (Holt et al. 2015)

Table 13. Identities of potential entomopathogenic bacteria based on BLAST results

Saturnid Host	Sample codes	Species	Similar GenBank sequences (% similarity)
<i>Gonimbrasia zambesina</i>	LS20, 37, 47, 51, 52, 56, 58 & 61	<i>Enterococcus mundtii</i>	AB831185.1 (98-99%)
<i>Gonimbrasia zambesina</i>	LS32	<i>Bacillus cereus</i>	MH041186.1 (99%)
<i>Gonimbrasia zambesina</i>	LS23	<i>Staphylococcus sciuri</i>	MK205162.1 (99%)
<i>Gonimbrasia zambesina</i>	LS31 & 38	<i>Staphylococcus gallinarum</i>	MH371285.1 (99%)
<i>Gonimbrasia zambesina</i>	LS62	<i>Pseudomonas putida</i>	JN679855.1 (99%)
<i>Gonimbrasia zambesina</i>	LS28, 29, 46, 48, 50, 53, 54, 64, 66 & 68	<i>Enterobacter hormaechei</i>	CP031726.1 (97%)
<i>Bunaea alcinoe</i>	LS75	<i>Enterococcus faecalis</i>	MF369863.1 (98%)
<i>Bunaea alcinoe</i>	LS71	<i>Staphylococcus sciuri</i>	MK205162.1 (99%)
<i>Bunaea alcinoe</i>	LS33	<i>Staphylococcus gallinarum</i>	MH371285.1 (98%)
<i>Bunaea alcinoe</i>	LS2	<i>Enterobacter hormaechei</i>	CP031726.1 (97%)
<i>Gonimbrasia krucki</i>	LS39	<i>Staphylococcus gallinarum</i>	MH371285.1 (99%)

<i>Gonimbrasia cocaulti</i>	LS4	<i>Alcaligenes faecalis</i>	MK517568.1 (98%)
<i>Gonimbrasia cocaulti</i>	LS8	<i>Stenotrophomonas maltophilia</i>	MK300721.1 (98%)
<i>Gonimbrasia cocaulti</i>	LS55	<i>Enterobacter hormaechei</i>	CP031726.1 (97%)

5.4.3 Fungi

Ten fungal isolates were isolated from three species of saturniid larvae. *Phoma* sp., *Chaetomidium* sp. and *Mucor circinelloides* were isolated from *B. alcinoe* while *Curvularia prasadii* and *Sordaria* sp. were isolated from *Go. krucki*. *Pestalotiopsis disseminata*, *Penicillium citrinum*, *Neopestalotiopsis* sp and *Nigrospora* sp. were isolated from *Go. zambesina*. All of them are potential entomopathogens except *Sordaria* sp. and *Neopestalotiopsis* sp. (Table 14).

Table 14. Fungi isolated from dead saturniid larvae

Fungus (Identified using NCBI-BLAST)	Number of isolates	Saturniid host	Potential Entomopathogen
<i>Phoma</i> sp.	1	<i>B. alcinoe</i>	Yes (Gouli et al., 2013)
<i>Chaetomidium</i> sp.	1	<i>B. alcinoe</i>	Yes (Arzanlou et al. 2012)
<i>Mucor circinelloides</i>	1	<i>B. alcinoe</i>	Yes (Shan et al. 2019; Mathai et al.1990)
<i>Curvularia prasadii</i>	1	<i>Go. krucki</i>	Yes (Sharma et al., 2012)
<i>Sordaria</i> sp.	1	<i>Go. krucki</i>	No
<i>Pestalotiopsis disseminata</i>	6	<i>Go. zambesina</i>	Yes (Lv et al. 2011)
<i>Penicillium citrinum</i>	1	<i>Go. zambesina</i>	Yes (Maketon et al. 2014)
<i>Nigrospora</i> sp.	1	<i>Go. zambesina</i>	Yes (Thakur et al. 2012)
<i>Neopestalotiopsis</i> sp.	2	<i>Go. zambesina</i>	No
<i>Nigrospora</i> sp.	1	<i>Go. zambesina</i>	Yes (Thakur et al. 2012)

5.4.4 Life cycle

Three generations of *Go. zambesina* and *C. forda* were successfully raised in the laboratory by feeding on their respective food plants. Life cycle data for three generations was recorded for both *Go. zambesina* and *C. forda*. The complete life cycle of *Go. zambesina* was 152- 170 days (Figure 19). For *G. zambesina*, the male moths emerged earlier than female moths by 2-10 days (Figure 20).

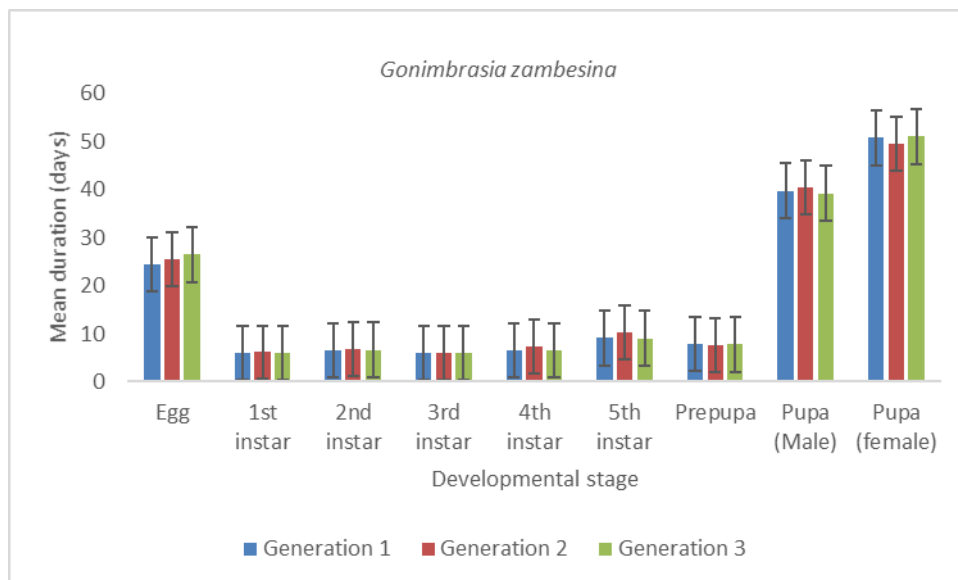


Figure 19. Life cycle of *Gonimbrasia zambesina*

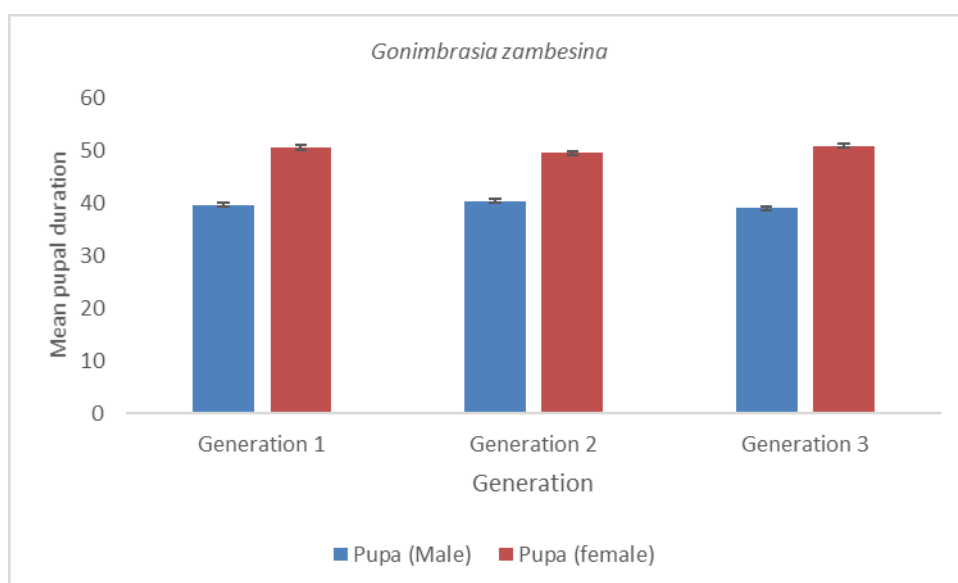


Figure 20. Pupal duration comparison for three generations of *Gonimbrasia zambesina*

For *C. forda*, we recorded three generations and replicated once. The complete lifecycle of *C. forda* from egg to adult stage was between 102- 246 days (Figure 21). Larvae took 5-7 days to moult to the next instar. *Cirina forda* male moths emerged earlier than female moths by 3-9 days (Figure 22).

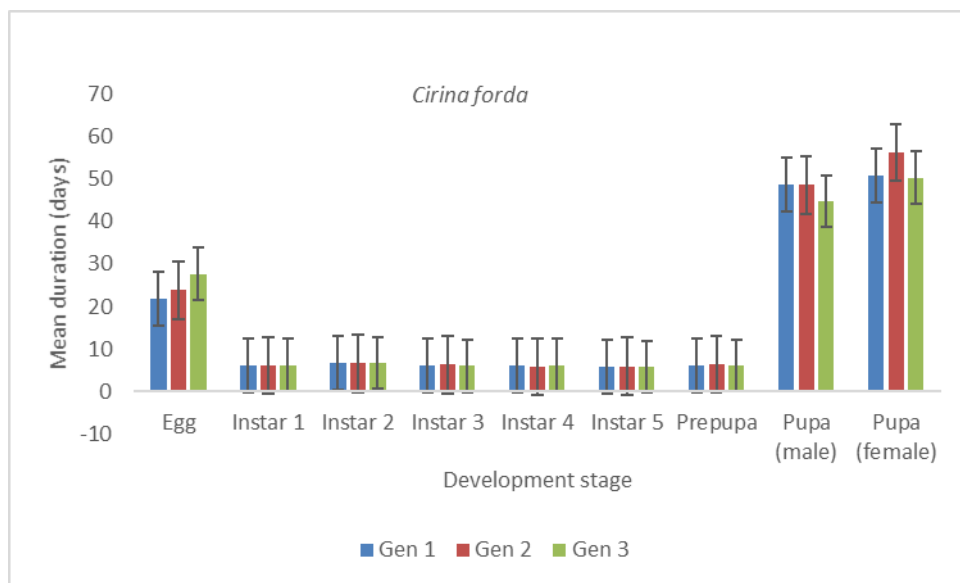


Figure 21. Life cycle of *Cirina forda*



Figure 22. Life cycle of *Cirina forda* (replication 1)

5.5 Discussion

From our findings, edible saturniids are attacked by parasitoids belonging to the orders Diptera and Hymenoptera. It is evident that parasitoids play a role in the natural regulation of edible saturniid populations since they affect the eggs, larvae and pupae. In this study, Chalcid wasps belonging to the subfamily Entedoninae (Eulophidae) were observed in *B. alcinoe* eggs. However, there are no records of parasitoids belonging to the subfamily Entedoninae parasitizing *B. alcinoe* eggs. This is also the first report of *Eupelmus* sp. (Eupelmidae) parasitizing *Go. krucki* eggs. *Eupelmus urozonus* has been recorded as a parasitoid of *G. belina* eggs (Van Den Berg 1974). Dipterans belonging to the Tachinidae, Ichneumonidae and Sarcophagidae families were the major larval-pupal parasitoids of saturniids in this study. A *Sarcophaga* sp. emerged from *B. alcinoe* pupae. This parasitoid has been reported before in *B. alcinoe* in Nigeria (Akanbi 1973). An undescribed species of *Ceromyia* was recorded from *G. belina*, *G. maja* and *B. alcinoe*. Other Tachniids observed in *G. zambesina* and *C. forda*. Tachniids have been previously recorded in edible saturniids including *G. belina* (Ditlhogo 1996), *B. alcinoe*, and *Cirina forda* (Akanbi 1973; Van Den Berg 1974). A larval-pupal parasitoid *Euryophion pisinnus* (Gauld & Mitchell) was observed on *B. alcinoe*. Previously, other Ichneumonids including *Euryophion latipennis* and *Encardia picta* have been observed emerging from *B. alcinoe* pupae. *Euryophion nigripennis* has also been recorded in *G. belina* (Gauld and Mitchell 1978; Oberprieler 1990). Larval parasitoids constituted braconids. *Glyptapanteles maculitarsis* (Cameron) was recorded in *B. alcinoe*. This finding concurs with other studies where *Glyptapanteles maculitarsis* parasitized *Gonimbrasia cytherea*, *G. zambesina*, *Gonimbrasia tyrreha*, *B. alcinoe* and *G. maja* larvae (Geertsema 1975; Van Den Berg 1971; Walker et al. 1990). *Aleiodes trifasciatus* (Enderlein) recorded in this study has not been observed before as a parasitoid of *G. zambesina*. *Cotesia* sp. found parasitizing *G. zambesina*, *G. belina* and *B. alcinoe* larvae has also not been reported before in these saturniids. All the fungi isolated were potentially entomopathogenic except *Sordaria* sp. and *Neopestalotiopsis* sp. Despite being plant pathogens, metabolites of *Nigrospora* species have been found to be entomopathogenic (Thakur et al. 2012). While nine

potentially entomopathogenic bacteria species were isolated from different saturniid species, their pathogenicity on the saturniids was not confirmed.

Moths lived for 2-5 days, they lacked mouth parts to feed and therefore mated, laid eggs and died. Male moths emerged earlier than female moths for both insects. Owing to this short adult longevity and differences in emerging times, both *G. zambesina* and *C. forda* colonies could not expand since the moths died before they could mate.

In DRC, locals have carried larvae of *C. forda* from the forests and placed them on trees near the homestead (Latham 2015). This could be exploited in future in a semi-wild mass rearing arrangement where locals plant host plants near their homesteads and inoculate them with saturniid eggs collected in the wild.

We tried to feed the larvae of *G. zambesina* on a semisynthetic diet but the larvae died at second instar stage. The biggest challenge was presenting the diet in a form that mimics leaves in the wild.

5.6 Conclusion

Egg, larval and pupal parasitoids of edible saturniids in Kenya were successfully identified. We have reported for the first time a *Eupelmus* sp. (*Eupelmidae*) and Entedoninae (*Eulophidae*) parasitizing *Go. krucki* and *B. alcinoe* eggs respectively. A new species of *Ceromyia* was observed as a larval-pupal parasitoid of *Go. belina*, *G. maja* and *B. alcinoe*. These parasitoids may contribute to the reduction of edible saturniid populations in the wild. While potentially entomopathogenic bacteria and fungus were isolated in edible saturniid samples, the analyses were not conclusive that they are pathogenic in these saturniids. Mass rearing of edible saturniids is still necessary in order to protect wild habitats. Semi-wild rearing is recommended since it encourages planting more trees (host plants). More studies are required to understand and break diapause at egg and pupa stage of edible saturniids.

6.0 General discussion

The first objective of this study aimed at understanding community perceptions on insect consumption in Kenya. Insect consumption has been previously reported in Kenya (van Huis 2019), though consumption of saturniids has not been well documented. We report for the first time, the consumption of *C. forda* and *G. zambesina* in the country. These saturniid caterpillars are mainly consumed by the Giriama community from coastal Kenya. We found termites to be the most and lake flies the least consumed insects in Kenya. We attribute these findings to availability of the insects in the study areas. For instance, lake flies can only be found near lake water bodies (Ayieko and Oriaro 2008) hence their consumption in the Lake Victoria region in Western Kenya. Previous studies show that insect consumption can be influenced by different factors including seasonality, availability and ethnicity (van Huis 2013; Kinyuru et al. 2013). Saturniid caterpillars are consumed by 8% of the respondents in this study and the consumption is confined within the Giriama community which is most likely attributable to culture and ethnicity.

We further sought to understand why people did not consume insects. Respondents cited disgust and fear of insects as the main reasons they did not consume insects. Although we reported more consumers than non-consumers, there is a marked reduction in the frequency of insect consumption in Kenya. Younger respondents aged 30 years and below had a lower insect consumption rate compared to their older counterparts. This could be attributed to modernisation among the youth, a knowledge gap between the youth and older people regarding insect consumption and lack of availability of edible insects (Dube et al. 2013; Obopile and Seeletso 2013; Riggi et al. 2016). This is in contrast to findings in Ivory Coast where more young people are reported to consume insects (Ehounou et al. 2018). Insects are also perceived as food for the poor (Tao and Li 2018) which could explain the reduction in insect consumption among younger generations. Limited availability could be attributed to increase in agriculture that often encroaches into the natural habitats of edible insects, hence interfering with their natural populations. Insect farming could be a possible solution here.

We reported no close association between insect consumption and education. However, a study in Zimbabwe showed that educated people engaged less in insect consumption (Manditsera et al. 2018) with more farmers than civil servants inclined to

consume insects. This is probably due to modernisation and the city lifestyle where most civil servants are found. In this study, more women consumed insects than men. However, insect collection was mostly carried out by children while selling was dominated by women. Similar findings have been recorded for the collection and trading of mopane worms in southern Africa (Stack et al. 2003). Insect collection and trading is not considered a main source of income by Kenyan men so they shy away from such activities

While saturniids in Kenya are consumed primarily in the coastal region, respondents from other parts of the country consider them often as harmful and poisonous. More than 30% of the respondents were willing to consume saturniid caterpillars as long as they were processed into more palatable forms. People in Western Kenya prefer biscuits made of cricket flour compared to whole crickets (Münke-Svendsen et al. 2016). Processing edible saturniids into powders incorporated in other foods could possibly increase their consumption in Kenya. Increasing awareness on the benefits of insect consumption could also promote entomophagy. While respondents were interested in saturniid rearing, they cited income generation as their main motivation. Yet many were concerned with the frequent lack of ready markets for the insects if they opted to rear them.

The next aim of this study was to document edible saturniid diversity in Kenya, their distribution and their host plants. Such information is important to promote their consumption and conservation. We carried out both morphological and molecular characterization of saturniids. We documented seven species of saturniids, among them, three are consumed in Kenya, namely: *C. forda*, *Go. zambesina* and *B. alcinoe*. These saturniids are also widely consumed in other parts of the continent including West, Central and Southern Africa (Badanaro et al. 2014; Kachapulula et al. 2018; Mabossy-Mobouna et al. 2016).

We noted morphological differences among insects of the same species. *Bunaea alcinoe* larvae found in Kenya were black, while a sample collected in Nigeria was red in colour. We report for the first time, a different genetic configuration between the two morphotypes. Although both colour forms have been cited feeding on *Cananga odorata* (Lam.) Hook.f. & Thomson (*Annonaceae*) Latham 2015), more studies on their mating compatibility and molecular differences are required for clarity on their

taxonomy. *Gonimbrasia zambesina* moths and larvae exhibited two colour forms. Black spined larvae resulted in green moths while red spined larvae resulted in brown ones. The green form had previously been reported as *Go. zambesina* (Pinhey 1956; Goff 2019) while the brown form had inconclusively been reported as *Gonimbrasia said* (Pinhey 1956). In our study, we could conclusively document both forms as genetically identical and as *Gonimbrasia zambesina*.

We further investigated the temporal distribution of the two saturniids (*C. forda* and *B. alcinoe*). Occurrence of their larvae coincided with the rainy seasons, i.e. April-June and October-December. However, in Togo, *C. forda* larvae appear only once a year between July and September (Badanaro et al. 2014) and similarly in DR Congo once between November and January (Latham 2015). *Bunaea alcinoe* is also univoltine in DR Congo and occurs between October and May (Latham 2015).

To further understand and predict their distribution, we generated a habitat suitability model of the *C. forda* and *B. alcinoe*. The model demonstrated that both species thrive in the tropical regions of Africa with *B. alcinoe* slightly spreading into the southern region of the continent. Our findings concur with previous reports on availability and consumption of the two species in southern, central and western Africa (Badanaro et al. 2014; Dwomoh et al. 2010; Kelemu et al. 2015; Kwiri et al. 2020; Mabossy-Mobouna et al. 2016; Mbata et al. 2002). Unfortunately, our future predictions show a reduction in habitat suitability for the two species emphasizing the need for conservation of these edible saturniids and their natural habitats. In terms of their potential for human consumption due to their endangered presence and their strong seasonality there is clearly a need to develop suitable mass production techniques for the insects.

We further characterized the host plants of the identified saturniids. They feed on specific host plants whose availability determines the distribution of saturniids. Previous studies indicated a wide range of host plants for *B. alcinoe* (Akanbi 1973; Latham 2015). However, for Kenya we found *B. alcinoe* only on *B. aegyptica* and *B. glabra*. Both tree species have been recorded as host plants for *B. alcinoe* in Nigeria as well (Akanbi 1973). Contrary to *B. alcinoe*, *C. forda* has been thought to possess a much more limited diversity of host plants (Badanaro et al. 2014; Dwomoh et al. 2010; Latham 2015). Yet, we report for the first time, *E. divinorum* Hiern (Ebenaceae), *A.*

mearnsii De Wild (Fabaceae) and *Manilkara sulcata* (Engl.) Dubard (Sapotaceae) as host plants of *C. forda*. In DR. Congo, it mainly feeds on *Crossopteryx febrifuga* (Latham 2015), while in West Africa it prefers the shea butter tree, *Vittelaria paradoxa* (Badanaro et al. 2014; Dwomoh et al. 2010) and the latter explains why it is commonly known as the shea caterpillar. We collected *Gonimbrasia zambesina* larvae from mango and cashew nut trees which have been previously reported as host plants (Pinhey 1956). All these host plants have other uses in the communities. For example, mango and cashew are domesticated fruit trees while *E. divinorum*, *V. tortilis* Forssk (Fabaceae) and *V. nilotica* (Fabaceae) are a source of firewood among other uses (Bussmann et al. 2006; Orwa et al. 2009; Tian 2017). Such knowledge can be leveraged on to encourage communities to plant these host plants and conserve them.

We also sought to demystify the decimating biotic factors that affect saturniid populations in the wild. We focused on parasitoids and potential entomopathogens. An undescribed chalcid sp. belonging to the family Entedoninae and an undescribed Eupelmidae were observed in *B. alcinoe* and *Go. krucki* eggs. Tachinids, ichneumonids and sarcophagids proved to be the major larval-pupal parasitoids of saturniids in our study. In terms of larval parasitoids we found predominantly braconids like *Glyptapanteles maculitarsis* (Cameron) in *B. alcinoe*. *Aleiodes trifasciatus* (Enderlein) was for the first time observed as a parasitoid of *G. zambesina* and similarly a *Cotesia* sp. found parasitizing *G. zambesina*, *G. belina* and *B. alcinoe* larvae had not been known as a parasitoid of these saturniids.

We isolated eight potentially entomopathogenic fungi and nine potentially entomopathogenic bacteria. However, further experiments on their pathogenicity for saturniids could not be carried out due to lack of insect colonies in the laboratory. Yet we were able to establish life cycles of *C. forda* and *G. zambesina*, though maintaining a colony in the laboratory proved to be difficult since the moths emerged at different times and died before they could mate. Semi-wild farming which involves incubating saturniid eggs on host plants and using netting to protect them from parasitoids is a proven practice, as for example with *C. forda* by local farmers in DR Congo (Latham 2015).

7.0 Conclusions and recommendations

7.1 Conclusions

Edible insects are consumed in Kenya, with termites and lake flies as the most and least popular species. Consumption of saturniid caterpillars is confined to the coastal region of the country, and particularly practiced by the Giriama community. Saturniid caterpillars are harvested exclusively in the wild.

Edible saturniids occur throughout Kenya, and different species have been documented in this research. They feed on specific host plants whose availability influences their distribution. Edible saturniids are seasonal, mostly appearing during and after the rainy season in Kenya and beyond. According to our model estimates, climate change might cause a slight reduction in the distribution of *C. forda* and *B. alcinoe* in Africa by the year 2050.

Edible saturniids are attacked by different parasitoids at the egg, larval and pupal stage. More detailed knowledge of these parasitoids will inform measures to prevent infestation during mass rearing. However, for mass rearing the main challenge is pupal and egg diapause, which also occurs in the wild, and so far has prevented the development of suitable mass rearing techniques for edible saturniids in captivity. Additional challenges are lack of synchronisation of male and female moth emergence and the lack of semisynthetic diets in a form that mimics plant leaves that edible saturniid caterpillars feed on in the wild.

7.2 Recommendations

We noted a decrease in insect consumption among young people. Hence the need to create greater awareness on the benefits of consuming insects. This should be accompanied by research on ways how to process edible insects into more palatable forms. While edible saturniids in Kenya are consumed mainly by the Giriama community, they widely occur in the eastern part of the country and the Lake Victoria region where they are presently not consumed by the locals despite the history of entomophagy in these regions. Sensitizing the communities on the benefits associated with the practice might spur consumption of saturniids and other edible insects. Because of the strong seasonality of edible saturniid caterpillars in Kenya, we recommend further research on mass rearing. A semi-wild set-up should be considered whereby communities plant host trees near their homesteads and

inoculate them with saturniid eggs collected in the wild. Instead of the current practice of saturniid caterpillars collection in the wild, we advocate for conservation of natural habitats by legislating protection of their host plants. Sensitization on the effects of deforestation and overharvesting will help the community conserve edible saturniid species and their natural habitats, especially if this is accompanied by training on techniques of semi-wild mass rearing. A crucial prerequisite for such techniques to be effective is the development of methods to prevent parasitoid infestations of the rearing setup. Our findings on life cycle and diapause in selected edible saturniids could prove helpful in this development process.

While we documented the occurrence of saturniid caterpillars in Kenya, their distribution, host plants and consumption, further research is required to analyse the economic benefits of edible saturniids within the Giriama community and beyond.

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9.0 APPENDICES

Appendix 1: Edible saturniids in Africa and their host plants

Insect	Countries consumed	Host plants	References
<i>Cirina forda</i>	Togo, Nigeria DRC, Zambia, South Africa, Botswana, Burkina Faso, Mozambique, Namibia, Ghana, Chad, Zimbabwe, Cameroon	<i>Vitellaria paradoxa</i> , <i>Crossopteryx febrifuga</i> , <i>Burkea africana</i> , <i>Erythrophleum suaveolens</i> , <i>Baillonella toxisperma</i>	(Akanbi 2002; Badanaro et al. 2014; Kelemu et al. 2015; Kozanayi and Frost 2002; Latham 2015; Ngute et al. 2020)
<i>Gynanisa maja</i> Strand	Zambia, Zimbabwe, South Africa, Botswana, Malawi, Namibia, Angola	<i>Colophospemum mopane</i> , <i>Julbernardia paniculate</i> , <i>Isobertlinia angolensis</i> , <i>Bachystegia glaberrima</i> , <i>Diplorhynchus condilocarpon</i> , <i>Burkea africana</i>	(Dube et al. 2013; Kozanayi and Frost 2002; Mbata and Chidumayo 2003; Nonaka 2009; Obopile and Seeletso 2013)
<i>Gonimbrasia belina</i>	Zambia, Zimbabwe, DRC, South Africa, Botswana, Malawi, Namibia, Angola	<i>Colophospemum mopane</i> , <i>Julbernardia paniculate</i> , <i>Isobertlinia angolensis</i> , <i>Bachystegia glaberrima</i> , <i>Diplorhynchus condilocarpon</i>	(Kelemu et al. 2015; Kozanayi and Frost 2002; Obopile and Seeletso 2013; Siulapwa et al. 2014; Thomas 2013)
<i>Bunaea alcinoe</i>	DRC, Zambia, South Africa, Cameroon, Congo, Central African Republic (CAR), Zimbabwe, Nigeria, Tanzania	<i>Parkia biglobosa</i> , <i>Proposis africana</i> , <i>Khaya senegalensis</i> , <i>Gmelina arborea</i> , <i>Spondias mombim</i> , <i>Pentaclethra crophylla</i> , <i>Terminia cattapa</i> , <i>Cananga odorata</i> , <i>Harungana madagascariensis</i> , <i>Sarcocephalus latifolius</i> , <i>Acacia auriculiformis</i> , <i>Mangifera indica</i> , <i>Dacryodes edulis</i> , <i>Crossopteryx febrifuga</i> , <i>Anthocleista schweinfurthii</i> , <i>Erythrophleum suaveolens</i> , <i>Amphimas ferrugineus</i> , <i>Holarrhena floribunda</i> , <i>Pyrranthus Congolensis</i> , <i>Petersianthus macrocarpus</i> , <i>Discoglyptemna calonecira</i> , <i>Funtumia Africana</i> , <i>Ricinodendron heudelotii</i> , <i>Cussonia barterri</i> , <i>Balanites aegyptica</i> , <i>Fagraea fragrans</i> , <i>Spondias mombin</i> , <i>Cleistopholis patens</i> , <i>Ekebergia senegalensis</i>	(Akanbi 1973; Amadi et al. 2005; Fasoranti and Ajiboye 1993; Kelemu et al. 2015; Latham 2015; Mabossy-Mobouna et al. 2016; Ngute et al. 2020)
<i>Gonimbrasia alopia</i>	Cameroon, Congo	<i>Mangifera indica</i> , <i>Dacryodes edulis</i>	(Mabossy-Mobouna et al. 2016; Ngute et al. 2020)
<i>Gonimbrasia epimethea</i>	DRC, Zambia, South Africa, Cameroon,	<i>Erythrophleum suaveolens</i> , <i>Amphimas ferrugineus</i> , <i>Pyrranthus Congolensis</i> , <i>Petersianthus macrocarpus</i> , <i>Discoglyptemna calonecira</i> , <i>Funtumia Africana</i> ,	(Akanbi 1973; Kelemu et al. 2015; Latham 2015; Mbata and Chidumayo 2003; Ngute et al. 2020)

	Congo, CAR, Zimbabwe	<i>Ricinodendron heudelotii</i> , <i>Dacryodes edulis</i> , <i>Holarrhena floribunda</i> , <i>Albizia ferruginea</i> . <i>Holarrhena floribunda</i> , <i>Acacia auriculiformis</i> , <i>Ekebergia senegalensis</i>	
<i>Gonimbrasia oyemensis</i>	Cameroon, DRC, CAR, Congo	<i>Maesopsis eminii</i> , <i>Entandrophragma cylindricum</i> <i>Pentaclethra macrophylla</i> , <i>Macaranga monandra</i> , <i>Albizia ferruginea</i> , <i>Acacia auriculiformis</i>	(Latham 2015; Mabossy-Mobouna et al. 2016; Ngute et al. 2020)
<i>Cirina butyrospermi</i>	Burkina Faso, Nigeria, Mali, Ghana	<i>Vitellaria paradoxa</i>	(Anvo et al. 2016; Yapo et al. 2017)
<i>Lobobunaea phaedusa</i>	DRC	<i>Annona senegalensis</i> , <i>Crossopteryx febrifuga</i> .	(Latham 2015)
<i>Gonimbrasia eblis</i> Strecker	DRC	<i>Chaetocarpus africanus</i> , <i>Manotes expansa</i> , <i>Ochna afzelii</i> , <i>Mangifera indica</i> , <i>Acacia auriculiformis</i>	(Latham 2015)
<i>Gonimbrasia obscura</i>	DRC, Cameroon	<i>Pentaclethra macrophylla</i> , <i>Macaranga monandra</i> , <i>Albizia ferruginea</i> , <i>Acacia auriculiformis</i> , <i>Dacryodes edulis</i> , <i>Bridelia micrantha</i> , <i>Margaritaria discoidea</i> , <i>Trema orientalis</i>	(Latham 2015; Ngute et al. 2020)
<i>Gonimbrasia wahlbergii</i> Druce	DRC	<i>Maesobotrya vermeulenii</i>	(Latham 2015)
<i>Gonimbrasia alopia</i> Westwood	DRC, Cameroon	<i>Albizia ferruginea</i> , <i>Manotes expansa</i> , <i>Millettia barteri</i> , <i>Chaetocarpus africanus</i> <i>Mangifera indica</i> , <i>Dacryodes edulis</i>	(Latham 2015; Ngute et al. 2020)
<i>Gonimbrasia anthina</i> Karsch	DRC	<i>Aframomum alboviolaceum</i> , <i>Antidesma venosum</i> , <i>Strychnos pungens</i> , <i>Manotes expansa</i> .	(Latham 2015)

Appendix 2: Local names of edible saturniids in Africa.

Species	Country	Local name	Ethnic group
<i>B. alcinoe</i> , <i>G. alopia</i> , <i>G. eblis</i> , <i>G. Obscura</i>	Congo	Binkele	Kongo,Lari
<i>B. alcinoe</i> , <i>G. alopia</i> , <i>G. eblis</i> <i>G. oyemensis</i> , <i>G. Obscura</i>	Congo	Inkele	Mbere, Mbosi, Northern Teke
<i>B. alcinoe</i> , <i>G. alopia</i> , <i>G. eblis</i>	Congo	Inkele	Teke
<i>B. alcinoe</i> , <i>G. alopia</i> , <i>G. anthinoides</i>	Congo	Mposo	Western Teke
<i>B. alcinoe</i>	Nigeria	Igu	Igbo
<i>B. alcinoe</i>	CAR	Selainbetoyo	Gbaya Bodoë
<i>B. alcinoe</i>	DRC	Makedikedi	Kikongo
<i>B. alcinoe</i>	DRC	Aisoalima	Bole
<i>B. alcinoe</i>	DRC	Baisobilo	Topoke
<i>B. alcinoe</i>	DRC	Finakifumbe	Chibemba
<i>B. alcinoe</i>	DRC	Mubamangoma	Katanga
<i>B. alcinoe</i>	Zambia	Chifumbe	Kilamba
<i>B. alcinoe</i>	Zambia	Muhwititi	Kilumba
<i>B. alcinoe</i>	Botswana	Phata	Setswana
<i>G. alopia</i>	DRC	Misongo	Kongo
<i>G. alopia</i>	DRC	Malemba	Kongo
<i>G. alopia</i>	DRC	Baetsuka	Mbanza
<i>G. eblis</i>	DRC	Nkankah	Western Teke
<i>G. oyemensis</i>	DRC	Mboyoy	Baaka, Bomitaba, Kaka, Bodongo, Bonguili, Mbonjo, Yasswa
<i>G. oyemensis</i>	DRC	Isie, Esie	Bakouele
<i>G. oyemensis</i> , <i>G. epimethea</i> , <i>G. obscura</i>	DRC	Mbindzu	Bobangi, Likuba, Moi
<i>G. oyemensis</i> , <i>G. epimethea</i> , <i>G. obscura</i>	DRC	Misie	Djem
<i>G. oyemensis</i> , <i>G. epimethea</i> , <i>G. obscura</i>	DRC	Binkele	Kiri
<i>G. oyemensis</i> , <i>G. epimethea</i> , <i>G. obscura</i>	DRC	Mbindzo	Lingala, Bomwali, Bongili
<i>G. oyemensis</i>	DRC	Boyo	Mbendjele
<i>G. oyemensis</i>	DRC	Mboyoy, se	Pomo
<i>G. oyemensis</i>	DRC	Etob'etama	Western Teke

<i>G. oyemensis</i>	CAR	Mboyo	Bofi
<i>G. oyemensis, G. obscura</i>	DRC	Minsendi	Kongo
<i>G. oyemensis, G. obscura</i>	DRC	Liboyo	Lingala
<i>G. oyemensis, G. obscura</i>	DRC	E-ontokala	Ba-Twa
<i>G. oyemensis, G. obscura</i>	DRC	Bihoyo	Ngando
<i>G. epimethea</i>	Congo	Kuluka, Nzamba	Baka
<i>G. epimethea</i>	Congo	Epak, Ipak	Bakwele
<i>G. epimethea</i>	Congo	Embii	Bambamba
<i>G. epimethea, G. obscura</i>	Congo	Mbinzu	Bangi
<i>G. epimethea</i>	Congo	Kuhuka, Nkuluka	Bomitaba
<i>G. epimethea</i>	Congo	Kuluka	Bondongo
<i>G. epimethea</i>	Congo	Kulupa	Enyelle
<i>G. epimethea</i>	Congo	Kuluka	Kaka, Monzombo
<i>G. epimethea</i>	Congo	Mihuka	Kongo, Lari
<i>G. epimethea</i>	Congo	Koholi	Mbanza
<i>G. epimethea</i>	Congo	Mbindzi	Mbere, Mbosi, Northern Teke
<i>G. epimethea</i>	Congo	Mbizu	Ndasa
<i>G. epimethea</i>	Congo	Pisi	Pomo
<i>G. epimethea</i>	Congo	Mobii	Western Teke
<i>G. epimethea</i>	Congo	Pusu	Yasswa
<i>G. epimethea</i>	Congo	Mepah	Djem
<i>G. epimethea</i>	Congo	Nkuluka	Mbonjo
<i>G. epimethea</i>	CAR	Sounga	Bofi
<i>G. epimethea</i>	DRC	Pamba, Mishila	Bemba
<i>G. epimethea</i>	DRC	Mvinsu	Kongo
<i>G. epimethea</i>	DRC	Moilo	Twa
<i>G. epimethea</i>	DRC	Bafoyo	Komo
<i>G. epimethea</i>	DRC	Sogo	Topoke
<i>G. epimethea</i>	Zambia	Mishila	Lamba
<i>G. epimethea</i>	Zambia	Makomechina	Lunda
<i>G. epimethea</i>	Zambia	Mpambata	Bisa

<i>C. forda</i>	Congo	Ngbanda	Baaka, Monzombo, Mbendjele
<i>C. forda</i>	Congo	Eler, Ilir	Bakouele
<i>C. forda</i>	Congo	Ngwanda	Bomitaba, Kaka, Mbonjo, Pomo, Bomwali, Bondongo, Bongili, Yasswa
<i>Cirina forda</i>	Congo	Milun	Djem
<i>C. forda</i>	Congo	Mpuampula	Lari
<i>C. forda</i>	Congo	Ajoh gbangbana	Mbanza
<i>C. forda</i>	Congo	Ndzandzaba, Mpampala	Western Teke
<i>C. forda</i>	Burkina faso	Chitoumou	Dioula
<i>C. forda</i>	Burkina faso	Bobo	Bobo-Dioulasso
<i>C. forda</i>	Togo	Salantonda	Moba
<i>C. forda</i>	Nigeria	Kanni	Nupe, Yoruba
<i>C. forda</i>	Nigeria	Munimuni	Yoruba
<i>C. forda</i>	DRC	Ngala	Kongo
<i>C. forda</i>	DRC	Ndanda	Lingala
<i>C. forda</i>	DRC	Bihomi	Ngando
<i>C. forda</i>	DRC	Bolabda	Topoke
<i>C. forda</i>	DRC	Makoso	Bapende, Kitshok
<i>C. forda</i>	DRC	Mikoso	Bemba
<i>C. forda</i>	Zambia	Mukoso	Lamba
<i>C. forda</i>	Zambia	Masesi	Lunda
<i>C. forda</i>	Zambia	Fikoso	Bisa
<i>C. forda</i>	Zimbabwe	Harati	Shona
<i>C. forda</i>	Botswana	Nato	Setswana
<i>G. obscura</i>	Congo	Kenakene	Baaka
<i>G. obscura</i>	Congo	Daswah	Bakwele
<i>G. obscura</i>	Congo	Matsentsene, Mankenkene	Bomitaba
<i>G. obscura</i>	Congo	Makekene	Bomwali, Bongili
<i>G. obscura</i>	Congo	Mankenkene	Bodongo, Enyelle
<i>G. obscura</i>	Congo	Dzaswom	Djem
<i>G. obscura</i>	Congo	Gengene	Kaka
<i>G. obscura</i>	Congo	Mbindzu	Likwala

<i>G. obscura</i>	Congo	Baladjah, Bladjah	Mbanza
<i>G. obscura</i>	Congo	Kenene	Mbedjele
<i>G. obscura</i>	Congo	Gegene	Mbonjo
<i>G. obscura</i>	Congo	Genegene	Monzombo
<i>G. obscura</i>	Congo	Mayulbatsie	Western Teke
<i>G. obscura</i>	CAR	Ngueguele	Bofi
<i>G. obscura</i>	CAR	Dok-kpare	Gbaya
<i>G. obscura</i>	DR Congo	Mo-pakala	Twa
<i>L. phaedusa</i>	Congo	Kungunu	Kongo, Western Teke
<i>L. phaedusa</i>	Congo	Mbaah	Western Teke
<i>L. phaedusa</i>	DRC	Kaba	Kongo
<i>G. belina</i>	Zambia	Mumpa	-
<i>G. belina</i>	Zimbabwe	Macimbi	Shona
<i>G. belina</i>	Botswana	Phane	Setswana
<i>G. maja</i>	Zambia	Chipumi	-
<i>G. maja</i>	Zimbabwe	Harati	Shona

Sources: (Anvo et al. 2016; Badanaro et al. 2014; Braide et al. 2011; Chavanduka 1975; Fasoranti and Ajiboye 1993; Latham 2003; Lisingo et al. 2010; Mabossy-Mobouna et al. 2016; Makhado et al. 2014; Mbata and Chidumayo 2003; Obopile and Seeletso 2013; Roulon-Doko 1998; Siulapwa et al. 2014; Temitope et al. 2014)

Appendix 3: Nutrition composition (%) of selected edible saturniids (based on dry matter)

Insect	Country	Fat	Crude Protein	Ash	Carbohydrate	Fibre	Reference
<i>C. forda</i>	Togo	-	52.39	16.48	-	8.92	(Badanaro et al. 2014)
<i>C. forda</i>	Nigeria	-	56.78	3.97	-	11.15	(Igbabul et al. 2015)
<i>C. forda</i>	Nigeria	-	54.36	3.76	-	11.03	(Igbabul et al. 2015)
<i>C. forda</i>	Nigeria	-	55.17	2.91	-	11.07	(Igbabul et al. 2015)
<i>C. forda</i>	Nigeria	16.12	31.4	7.62	7.8	10.8	(Paiko et al. 2014)
<i>C. forda</i>	Nigeria	14.3	74.35	3.1	2.36	6.01	(Agbidye et al. 2009)
<i>C. forda</i>	Nigeria	5.25	62.25	11.51	20.98	-	(Omotoso 2006)
		11.89	55.24	7.05	10.38	9.83	
<i>B. alcinoe</i>	Nigeria	10.85	44.23	5.92	-	11.8	(Dauda et al. 2014)
<i>B. alcinoe</i>	Nigeria	21.72	53.22	6.42	-	9.41	(Braide et al. 2010)
<i>B. alcinoe</i>	Nigeria	25	55	8	-	-	(Amadi et al. 2005)
<i>B. alcinoe</i>	Nigeria	14.1	74.34	2.85	3.16	5.55	(Agbidye et al. 2009)
		17.92	56.70	5.80	3.16	2.25	
<i>G. belina</i>	Zambia	10	56.95	7	7.8	-	(Siulapwa et al. 2014)
<i>G. belina</i>	Zimbabwe	16.4	55.4	8.3	8.2	16	(Siulapwa et al. 2014) (Musundire et al. 2016)
		13.2	56.18	7.65	8	16	
<i>G. maja</i>	Zambia	12.1	55.92	7.4	10.7	-	(Siulapwa et al. 2014)
<i>G. maja</i>	Zimbabwe	10.9	51.1	7.7	14.1	16.2	(Musundire et al. 2016)
		11.5	53.51	7.55	12.4	16.2	
<i>C. butyrospermi</i>	Burkina Faso	14.51	62.74	5.1	12.63	5.02	(Anvo et al. 2016)
<i>C. butyrospermi</i>	Ivory coast	-	55.41	4.89	-	2.68	(Yapo et al. 2017)
		14.51	59.08	5.00	12.63	3.85	

Appendix 4: Amino acid composition (mg/g) of selected edible saturniids (based on dry matter)

Insect	Country	His	Ile	Leu	Lys	Asp	Met	Cys	Gly	Pro	Phe+ Tyr	Thr	Val	Ala	Ser	Arg	Reference
<i>C. butyrospermi</i>	Ivory coast	25.6	26.4	27.4	52.5	0.0	6.9	3.0	31.0	29.4	53.3	37.0	43.4	32.4	22.1	67.4	(Yapo et al. 2017)
<i>C. butyrospermi</i>	Burkina Faso	28.0	31.1	40.8	61.3	72.8	15.8	12.4	26.3	31.8	50.0	23.1	52.2	55.9	16.6	48.6	(Anvo et al. 2016)
		26.8	28.8	34.1	56.9	72.8	11.4	7.7	28.7	30.6	51.7	30.1	47.8	44.2	19.4	58.0	
<i>C. forda</i>	Nigeria	17.1	24.6	33.4	29.5	65.6	9.7	8.0	29.7	30.1	-	49.2	55.3	49.6	32.2	65.4	(Paiko et al. 2014)
<i>C. forda</i>	Nigeria	25.7	32.7	75.1	56.4	86.5	23.5	14.5	38.0	31.2	-	41.8	31.8	43.1	30.3	67.8	(Igbabul et al. 2015)
<i>C. forda</i>	Nigeria	23.2	33.7	72.6	53.3	86.9	23.8	12.8	40.7	23.4	-	39.4	41.8	36.3	33.1	54.8	(Igbabul et al. 2015)
<i>C. forda</i>	Nigeria	24.0	43.1	65.5	62.4	94.3	22.2	10.7	27.2	27.4	-	32.2	40.5	43.4	29.1	50.2	(Igbabul et al. 2015)
		22.5	33.5	61.7	50.4	83.3	19.8	11.5	33.9	28.0	-	40.7	42.4	43.1	31.2	59.6	
<i>G. belina</i>	Zambia	18.4	13.0	18.3	25.6	31.3	4.1	1.1	17.9	18.6	35.8	18.4	19.1	23.6	17.5	45.7	(Siulapwa et al. 2014)
<i>G. belina</i>	Zimbabwe	15.0	21.5	31.2	35.8	53.0	10.0	10.4	22.6	22.6	56.3	27.4	27.5	25.2	27.1	28.5	(Glew et al. 1999)
		16.7	17.3	24.8	30.7	42.2	7.1	5.8	20.3	20.6	46.1	22.9	23.3	24.4	22.3	37.1	
<i>G. maja</i>	Zambia	25.3	18.8	27.2	40.2	39.9	8.2	2.2	19.9	25.0	61.5	22.6	20.9	25.5	23.1	31.4	(Siulapwa et al. 2014)
<i>B. alcinoe</i>	Nigeria	31.9	32.3	80.9	53.8	102.1	25.3	8.0	29.7	30.1	67.7	49.2	55.3	49.6	32.9	65.4	(Braide et al. 2010)
	Adult amino acid requirements	15	30	59	45		16	6			38	23	39				(WHO 2007)

Appendix 5: Mineral composition (mg/100g) of selected edible saturniids (based on dry matter)

Insect	Country	P	K	Ca	Mg	Fe	Mn	Cu	Zn	Na	Reference
<i>C. forda</i>	Togo	32.9	43.1	28.4	23.6	5.0	1.0	0.3	0.7	39.8	(Badanaro et al. 2014)
<i>C. forda</i>	Nigeria	213.5	64.0	33.2	62.3	5.3	1.1	-	3.8	45.3	(Omotoso 2006)
<i>C. forda</i>	Nigeria	111	65.0	32.2	67.3	12.9	7.5	-	3.7	45.5	(Paiko et al. 2014)
		119.1	57.4	31.3	51.0	7.8	3.2	0.3	2.8	43.5	
<i>B. alcinoe</i>	Nigeria	128.5	91.2	27	19.5	38.7	16.9	1.1	24.7	125.9	(Dauda et al. 2014)
<i>G. belina</i>	Zambia		10.2	127.8	69.7	26.7	1.5	0.3	-	42.1	(Siulapwa et al. 2014)
<i>G. maja</i>	Zambia		65.5	166.4	100	13.6	1.4	0.3	-	32.34	(Siulapwa et al. 2014)

Appendix 6: Table: Location and agro-ecological zones of the study sites in Kenya

Study site	GPS co-ordinates	Agro-ecological zone
Gilgil, Nakuru	0.4923° S, 36.3173° E	Zone III
Mbeere, Embu	0.5388° S, 37.4596° E	Zone II
Nkubu, Meru	0.0647° S, 37.6679° E	Zone II
Nanyuki, Laikipia	0.0074° N, 37.0722° E	Zone IV
Kasarani, Nairobi	1.2254° S, 36.8976° E	Zone II
Kibwezi, Makueni	2.4105° S, 37.9678° E	Zone V
Kishushe, Taita	3.3973° S, 38.5559° E	Zone V
Muhaka, Kwale	4.2879° S, 39.5653° E	Zone IV
Malindi, Kilifi	3.2192° S, 40.1169° E	Zone V
Mbita, Homabay	0.4368° S, 34.2060° E	Zone III
Mwingi, Kitui	0.9374° S, 38.0605° E	Zone V
Ngong, Kajiado	1.3562° S, 36.6688° E	Zone III
Matuu, Machakos	1.1407° S, 37.5481° E	Zone IV
Isiolo, Isiolo	0.3556° N, 37.5833° E	Zone IV
Makuyu, Murang'a	0.9026° S, 37.1875° E	Zone II

Appendix 7: Accession numbers of saturniid sequences submitted to the GenBank

	Sample	Accession number
1	SUB7133105 S17_Gzambesina_Kilifi	MT176774
2	SUB7133105 S18_Gzambesina_Kilifi	MT176775
3	SUB7133105 S79_Gzambesina_Kambiti	MT176776
4	SUB7133105 S95_Gzambesina_Embu	MT176777
5	SUB7133105 S97_Brown_Gzambesina_Makuyu	MT176778
6	SUB7133105 S98_Brown_Gzambesina_Makuyu	MT176779
7	SUB7133105 S99_Green_Gzambesina_Makuyu	MT176780
8	SUB7133105 S100_Green_Gzambesina_Makuyu	MT176781
9	SUB7133105 S101_Green_Gzambesina_Makuyu	MT176782
10	SUB7133105 S102_Green_Gzambesina_Makuyu	MT176783
11	SUB7133105 S104_Brown_Gzambesina_Makuyu	MT176784
12	SUB7133105 S105_Brown_Gzambesina_Makuyu	MT176785
13	SUB7133105 S107_Brown_Gzambesina_Makuyu	MT176786
14	SUB7133102 S48_Gufipana_Muhaka	MT176766
15	SUB7108149 S22_Gcocaulti_Matuu	MT159807
16	SUB7108149 S28_Gcocaulti_Matuu	MT159808
17	SUB7108149 S30_Gcocaulti_Mwingi	MT159809
18	SUB7108149 S32_Gcocaulti_Taita	MT159810
19	SUB7108149 S33_Gcocaulti_Taita	MT159811
20	SUB7108149 S34_Gcocaulti_Taita	MT159812
21	SUB7108145 IBB1_Gbelina	MT157403
22	SUB7108145 IBB2_Gbelina	MT157404
23	SUB7133102 S48_Gufipana_Muhaka	MT176766
24	SUB7074604 Nigeria1_Balcinoe	MT179695
25	SUB7074604 S84_Balcinoe_Matuu	MT179696
26	SUB7074604 S85_Balcinoe_Matuu	MT179697
27	SUB7074604 S87_Balcinoe_Nanyuki	MT179698

28	SUB7074604 S88_Balcinoe_Nanyuki	MT179699
29	SUB7074604 S90_Balcinoe_Mbita	MT179700
30	SUB7074604 S91_Balcinoe_Mbita	MT179701
31	SUB7074604 S92_Balcinoe_Embu	MT179702
32	SUB7074604 S93_Balcinoe_Embu	MT179703
33	SUB7074617 2CF_Cforda_Ngong	MT179704
34	SUB7074617 5CF_Cforda_Ngong	MT179705
35	SUB7074617 S6_Cforda_Mbita	MT179706
36	SUB7074617 S7_Cforda_Mbita	MT179707
37	SUB7074617 S54_Cforda_Gilgil	MT179708
38	SUB7074617 S55_Cforda_Kilifi	MT179709
39	SUB7138372 GM1_Gwestwoodi	MT182818
40	SUB7138372 GM1_Gwestwoodi	MT182818
41	SUB7138303 GMB2_Gnigra	MT179594
42	SUB7133097 S2_Gkrucki_Nairobi	MT178412
43	SUB7133097 S3_Gkrucki_Nairobi	MT178413

Appendix 8: Pairwise genetic distances of *Gonimbrasia zambesina* samples

	KAG94	EMG95	MAB97	MAB98	MAG99	MAG100	MAG101	MAG102	MAB104	MAB105	MAB106	MAB107	KFG17	KFG18	KAG79	SAPBA773-07	SAPBA772-07
KAG94	0.00%																
EMG95	0.00%	0.00%															
MAB97	0.15%	0.15%	0.00%														
MAB98	0.15%	0.15%	0.00%	0.00%													
MAG99	0.15%	0.15%	0.00%	0.00%	0.00%												
MAG100	0.15%	0.15%	0.00%	0.00%	0.00%	0.00%											
MAG101	0.30%	0.30%	0.15%	0.15%	0.15%	0.15%	0.00%										
MAG102	0.30%	0.30%	0.15%	0.15%	0.15%	0.15%	0.31%	0.00%									
MAB104	0.30%	0.30%	0.15%	0.15%	0.15%	0.15%	0.31%	0.31%	0.00%								
MAB105	0.15%	0.15%	0.00%	0.00%	0.00%	0.00%	0.15%	0.15%	0.15%	0.00%							
MAB106	0.15%	0.15%	0.00%	0.00%	0.00%	0.00%	0.15%	0.15%	0.15%	0.00%	0.00%						
MAB107	0.30%	0.30%	0.15%	0.15%	0.15%	0.15%	0.31%	0.31%	0.31%	0.15%	0.15%	0.00%					
KFG17	0.61%	0.61%	0.46%	0.46%	0.46%	0.46%	0.61%	0.61%	0.61%	0.46%	0.46%	0.61%	0.00%				
KFG18	0.61%	0.61%	0.46%	0.46%	0.46%	0.46%	0.61%	0.61%	0.61%	0.46%	0.46%	0.61%	0.00%	0.00%			
KAG79	0.15%	0.15%	0.00%	0.00%	0.00%	0.00%	0.15%	0.15%	0.15%	0.00%	0.00%	0.15%	0.46%	0.46%	0.00%		
SAPBA773-07	0.61%	0.61%	0.46%	0.46%	0.46%	0.46%	0.61%	0.61%	0.61%	0.46%	0.46%	0.61%	0.00%	0.00%	0.46%	0.00%	
SAPBA772-07	1.22%	1.22%	1.07%	1.07%	1.07%	1.07%	1.23%	1.23%	1.23%	1.07%	1.07%	1.23%	1.22%	1.22%	1.07%	1.22%	0.00%

Appendix 9: Pairwise genetic distances of *Bunaea alcinoe* samples

	NGR1	SATWA891-07.COI-5P	MAB84	MAB85	MAB86	NAB87	NAB88	MBB90	MBB91	EMB92	EMB93	LSAFR2238-12
NGR1	0.00%											
SATWA891-07.COI-5P	0.00%	0.00%										
MAB84	3.60%	3.60%	0.00%									
MAB85	3.44%	3.44%	0.15%	0.00%								
MAB86	3.44%	3.44%	0.15%	0.00%	0.00%							
NAB87	3.28%	3.28%	0.30%	0.15%	0.15%	0.00%						
NAB88	3.28%	3.28%	0.30%	0.15%	0.15%	0.00%	0.00%					
MBB90	3.44%	3.44%	0.15%	0.00%	0.00%	0.15%	0.15%	0.00%				
MBB91	3.45%	3.45%	0.46%	0.31%	0.31%	0.15%	0.15%	0.31%	0.00%			
EMB92	3.28%	3.28%	0.31%	0.15%	0.15%	0.00%	0.00%	0.15%	0.15%	0.00%		
EMB93	3.76%	3.76%	0.77%	0.61%	0.61%	0.46%	0.46%	0.61%	0.61%	0.46%	0.00%	
LSAFR2238-12	3.76%	3.76%	0.76%	0.61%	0.61%	0.76%	0.76%	0.61%	0.92%	0.76%	1.23%	0.00%