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Second Cycle Degree (MSc) in Sustainable Agriculture

# Worldwide Importance of Insect Pollination of Cucurbitaceae Crops: A Review

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## Abstract

Cucurbitaceae crops, including cucumber, melon, watermelons, squash and zucchini, are part of our modern diet worldwide. All Cucurbitaceae crops are cultivated for the fruit called 'pepo', ripe and unripe depending on the crop. Cucurbitaceae crops are highly dependent on pollinators because they are monoecious species with a separate male and female flower on the same plant. A systematic global literature review of 37 studies was conducted to study the effect pollination treatments have on Cucurbitaceae fruit quality. Most studies were performed during the 2000-2010 decade and the 2010-2020 decade, followed by the 1990-2000 decade, 2020-2021 and 1970s and 1980s. Over 38 fruit quality traits were looked for in each study with pollination treatments such as open, hand and/or closed. The results show that open pollination treatment versus the hand treatment and the exclusion treatment showed an improvement in the cumulative quality traits measured across studies. However, hand pollination in certain studies did show an improvement in quality traits. Native bees were sufficient to produce commercially acceptable fruits in most studies. Nonetheless, in order to have food security and sovereignty, pollination techniques need to be assessed depending on the agronomic, ecological, social and economic context surrounding the production of Cucurbitaceae crops.

## 1. Introduction

## 1.1. Importance of pollinators for plant, animal and human well being

Animal pollination is an essential, low cost and indispensable ecosystem service sustaining human well-fare, food security and crop cultivar diversity around the world (Aizen et al., 2009, Garibaldi et al., 2013, Klein 2007, Pardo 2020). Plant biodiversity and the succession of biological life depends on the effectiveness of plant and pollinator interaction occurring spontaneously and systematically in diverse landscapes (Kaiser-Bunbury et al. 2010, Steffan-Dewenter et al. 2002). Wild animal pollination is responsible for sexual reproduction, long-term agricultural production and genetic fitness of many crops. The interaction between plant and pollinator is probably one of the most important ecological and biological processes sustaining life (Ollerton et al. 2010).

Pollination not only ensures nutritious crops for human consumption, but a decline in pollination quantity and quality can mean a decline or extinction of most life. A decline in pollination services leads to a cascade effect in the biodiversity chain of plant and animal species (Biesmeijer 2006, Kearns et al. 1998). It is important to note that not all plant species require animal pollination in order to set seed, however, many plants with mixed mating systems still require animal pollination (Ollerton et al. 2011). Long-term selfing reproduction by individual plants with no genetic variations, compared to cross pollination, would end interbreeding. This eventually leads to an in-breeding genetic depression, and subsequent genetic poverty (Kearns 1998, Ollerton et al. 2010).

In order to maintain healthy ecosystems and healthy human populations with nutritious, low-cost food production, natural pollination services must be ensured. In the past decades, decline in pollinators has generated widespread concern in many national governments and non-government organizations (Ollerton 2010, Biesmeijer 2006, Kosior et al. 2007, Grixti et al. 2009, Colla and Packer 2008, Kearns et al. 1998), The disappearance or reduction in pollinator populations and diversity has serious consequences for human food security and sovereignty, through the loss of food webs and ecosystem functions.

Nonetheless, as Vanderbergen (2013) shows, the decline in pollinators is a complex and challenging ecological service to study with accuracy. Pollinator decline is a difficult phenomenon to pinpoint because species competitiveness with multiple biological interactions and increasing environmental changes, complicate the scenario by producing winners such as generalist pollinator species and affecting specialist pollinator species more. Another challenge in understanding pollinator-plant interaction, and the reason for this study, is scarce and isolated data on pollinator studies, which leaves scientists with incomplete conclusions and inferences. A systematic and global analysis of all the

research on pollinator populations' current situation needs to be established through continuous monitoring by researchers on mixed landscape matrices with agricultural and natural lands (Pardo et al., 2020).

#### 1.2. Landscape management, pollinators and agriculture

The conservation and sustainable management of natural areas, such as forests, native pastures, hedgerows and semi-natural forest, are key habitats for the breeding and fostering a diversity of wild pollinators (Steffan-Dewenter et al. 2002). Landscape management of natural areas has a direct and indirect effect on many ecosystem services for agriculture. Agricultural landscape simplification is responsible for the decline of functional biodiversity providing services to farmers such as pest biological control and pollination.

Despite its importance, the historical agriculture trend of the last century is of high production and landscape simplification, which negatively affects native pollinator populations (Garibaldi et al., 2013, Stokstad, E. 2007). Research in the last decades has shown a decline in the presence of insect pollinators. Additional challenges and pressures faced by pollinators include land-use changes, pesticide use, genetically modified crops, air pollution, climate change, and the invasion of alien pest species and diseases on the landscape (Potts et al., 2016; Vanbergen 2013).

One study in Italy at different landscape scales, showed that pollinator species richness tends to be lower in managed agricultural landscapes where pesticide use had more than one application (Brittain et al., 2010). This is the case particularly for neonicotinoid systemic pesticides which travel through the plant tissue and accumulate on the floral nectar and pollen of plants, causing damage and death to pollinator species (Stokstad, E. 2007).

Widespread concern on the decline in native pollinators abundance and diversity has been growing due to recent research (Biesmeijer et al. 2006, Kosior et al. 2007, Colla and Packer 2008, Grixti et al. 2009, Winfree et al. 2009). Pollination as an ecosystem service provides an increase in fruit and seed set of 39 out of the 57 major crops worldwide (Klein et al. 2007), providing an economic benefit of €153 billion annually, or 9.5% of the value of world agricultural production (Gallai et al. 2009). Other studies show that the global economic value of wild and managed pollination services was around \$215 billion in 2005.

The impact is even greater in the tropics where much of the world's biodiversity resides, and where animal pollinator dependence is the highest. According to Roubik (1995), 70% of all tropical crops show quality and quantity improvement due to animal pollination, meaning that 70% of the 124 of the crops used for human consumption require insect pollination. A loss in pollinators does not only mean a decline in food sources and diversity, but a reduction in plant based and plant derived medicine (Eilers et

al. 2011). Medicinal plants have been and continue to be used by native people around the world. This was recently the case during the Covid-19 pandemic. These plants are not only part of their cultural patrimony, but medicinal plants are the source for all of modern medicine. Furthermore, Aizen et al. (2009) have determined that cultivation of pollinator-dependent crops has, on average, increased in the last 50 years more than pollinator-independent crops, both in the developing and developed countries.

#### 1.3. Groups of Pollinators and their importance to crop quantity and quality

Social and solitary bees, wasps, flies, beetles, trips, butterflies, and moths comprise the vast majority of the world's pollinators, crucial for the pollination of fruit, vegetable, oil, seed, and nut crops (Free 1993, Vanbergen 2013). Worldwide there are over 20,000 species of bees, with flies and hoverflies as the second most frequent visitors to the majority of flowers with approximately 120,000 species. Finally, mammals and birds have also been reported to transfer pollen between flowers (Rhodes 2018). Most insects pollinate flowers accidentally as they search for food (nectar and pollen) at different stages of their development in order to satisfy their colony's needs.

Honey bees (*Apis mellifera*), might be the single most important pollinator species in the wild bees category. According to Oldroyd BP (2007) and Stockstad E. (2007) managed honey-bee hives have been in decline by almost 60% since the mid-1940s. This same phenomenon is happening across Western Europe, affecting a wide diversity of wild bees, primarily due to negatively managed landscape, pesticide use and habitat destruction. It has also been found that less mobile bee taxa were more affected by pesticide use than more mobile bees, primarily due to loss of surrounding semi-natural habitat (Steffan-Dewenter et al. 2002). Most research on pollinator loss focuses on bees, which suffer chronic exposure to a variety of stressors such as loss of abundance and variety of flowers, decline in suitable habitat, and long-term agrochemical exposure to pesticides, especially neonicotinoids (Rhodes 2018).

## 1.4. Cucurbitaceae crops and their economic global relevance

As mentioned above, the need for an abundance and diversity of pollinators is of undeniable importance for present and future human well-being. Since the beginning of the Holocene, some 12,000-10,000 years ago, humans began a gradual transition from hunting and gathering, to the domestication of wild plants. Their reliance on pollinators increased as they grew more and more dependent on them for fruit-trees and increasing use of arable crops.

The gourd family (*Cucurbitaceae*), which is grown in tropical and temperate climates, represents one of the many family crops domesticated. There are over 1000 species with 95 genera of Cucurbitaceae, from which 10 are of high economic value and consumption worldwide. These include cucumber (*Cucumis sativus*), bitter gourd (*Momordica charantia*), watermelon (*Citrullus lanatus*), preserving melon

(*Citrullus amarus*), honey melon (*Cucumis melo*), squash and zucchini (*Cucurbita pepo*), and bottle gourd (*Lagenaria siceraria*) (Chomicki et al. 2019). Other 23 varieties of Cucurbitaceae are of "minor" global economic importance, nonetheless they are of cultural value and represent an ecological niche for native and indigenous communities.

There are four major centers of cucurbit domestication: South and North America, Africa and Asia/Oceania, from which point they have been traditionally selected in Europe and reintroduced in other parts of the world. All the Cucurbitaceae crops are cultivated for the fruit called 'pepo', ripe and unripe depending on the crop. Some crops are used for their seeds and the oil pressed from them, some varieties are used for their fruit as a sponge, others for the sugary fruit used as a sweetener, and yet others for their medicinal properties. Most of the plants are climbers, having annual vines, and some are woody lianas, thorny shrubs and even trees. The stems are hairy and pentangular, with many species having large, yellow and white flowers.

Out of the family, most are annual or perennial herbs, and about half are of monoecious species and the other half dioecious species. Dioecious species have a sexual reproduction system where the male organ and female organ are on separate different plants. Monoecious species on the other hand, have both sexes on the same plant. In the case of some Cucurbitaceae crops, one flower is male and the other female within the same plant, from which the 'pepo' or fruit grows. Cucurbitaceae flowers are unisexual, with male and female flowers on different plants or on the same plant. Generally monoecious flowers are highly dependent on cross pollination.

The male flowers tend to open first, followed by the female flowers three to four days later. Usually, the male to female flower ratio is 3:1 male-female, but this can change depending on temperature characteristics. The male flower possesses both pollen and nectar, while the female flower only nectar. It is only when both the male and female flowers are open that pollination can occur. The female flower is open for only one day and is most receptive between the hours of 9 AM and 4 PM. For successful fruit production, flowers need to 1) have adequate amounts of fertilizers and water, and 2) the flowers require about 15 bee visits for maximum pollination.

Pumpkins, muskmelons, watermelons and cucumbers are warm weather plants, with a growth and yield quality best when days are warm and sunny. Squash and cucumbers usually require 50-65 days for the first production, while watermelon and muskmelon require 80-95 days (Basham C., Ells J., 1999). Several studies show that Cucurbitaceae, particularly watermelon and muskmelon, have an increase in yield and quality in relatively high plant densities (Edelstein and Nerson, 2002; Nerson 2002).

Cucurbitaceae are highly dependent on insect pollination since their pollen is not easily transferred from flower to flower by the wind due to its weight and stickiness. From all pollinators, honeybees are the most successful pollinators of Cucurbitaceae (Abu-Hammour & D. Wittmann, 2010, Cane et al., 2011,

Free 1993, Morse & Calderone 2000). Honeybees work the most intensively on flowers from around 6 AM to noon with the highest activity from 8-9 AM, visiting Cucurbitaceae crops mainly for their nectar. Other insects such as ants, wild bees and beetles are minor pollinators of Cucurbitaceae crops (Abu-Hammour & D. Wittmann, 2010). The aim of this study is to better understand the role insect pollination plays in Cucurbitaceae fruit quality worldwide.

## 2. Methodology

### 2.1. Literature Review and Data Collection

I performed a systematic review from May 2020 until December 2020, gathering studies in multiple scientific databases, such as ScienceDirect, Scopus, Web of Science, JSTOR, covering the time period of 1974 up to 2020. Some key words used for the search were: honey bee pollination quality AND quantity of zucchini; honey bee pollination quality AND quantity of pumpkin; apis pollination AND quality fruit; fruit quality AND insect pollination; honey bee pollination quality AND quantity of broccoli; pollination cucumber quality; pollination muskmelon quality; pollinator melon metabolites; pollinator quality fruit; pollinator sugar fruit; quality OR size OR sugars cucumber pollinator OR apis OR bumblebee OR bee OR honeybee OR pollination. Additional studies were included from the reference list of important research papers.

The pollination treatment ranged from hand pollination, to open pollination, as well as closed pollination, where pollinators were excluded. In some cases, open pollination meant that honey bees colonies were close by, but never the sole pollinators. Often open pollination involves the participation of wild honeybees, bumblebees, or native bees among other pollinators in each region.

#### 2.2. Data Analysis

The aim of the treatments was to compare pollination services of (1) open versus closed pollination, (2) hand versus open pollination, and (3) hand versus closed pollination. Following the same literature review method as Pardo (2020), each study was assigned one of the following codes based on the statistical tests for the differences performed by the original authors: Open > Exclusion = quality trait significantly greater in the open pollination, than the exclusion to pollinators treatment; Open < Hand = quality trait significantly greater in hand pollination treatment than open treatment; Hand > Exclusion = quality trait significantly greater in hand pollination treatment that exclusion treatment, and NS = no statistically significant difference between treatments. Hand pollination ranged from using cotton swabs, to growth regulators. When fruit quality traits were significantly different between open and closed, or hand and open, or hand and closed, each pollination treatment received a 1 if it had an effect, and 0 if it didn't have an effect or if the trait was not measured.

The qualitative fruit traits measured included weight, height, length, sugar content, mineral content, color, firmness, diameter, seed number, water content, among others. In order to obtain a better understanding of the pollinator groups, information of Cucurbitaceae crop pollinators was gathered across

all studies and graphically summarized. Additionally, a map of the countries from which the studies came was also performed, as well as the different time periods that the studies were gathered from.

## 3. Results

## 3.1. Number of studies across geographical regions and quality traits measured

The search concluded with over 40 studies analyzing the effects pollination had on Cucurbitaceae crops in 19 different countries. From the 42 articles, 5 were discarded as they did not show significance at 0.01 level, leaving a total of 37 studies to perform the literature review. From each study the following information was gathered: pollination methods used, fruit quality traits measured, pollinators involved, Cucurbitaceae crops studied, geographical location, and year of the study. A total of 178 trials were conducted across the 37 studies.

The majority of studies by far came from Asian countries (16), followed by North America (11), Africa (5), Europe (2), South America (2), and Oceania (1). However, the country with the most studies by far was USA (9), followed by India (5), Tanzania (3), Jordan (3), Brazil (2), Malaysia (2) and Canada (2). The rest of the countries, Ivory Coast, China, Greece, Japan, Poland, New Zealand, Pakistan, South Korea, Senegal, Egypt, Philippines and Thailand, performed only one study with our related topic (Fig. 1).

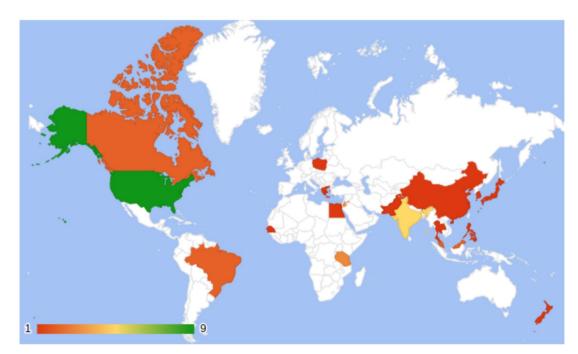


Fig. 1- Map showing distribution of studies looking at Cucurbitaceae crop pollination.

The frequency of the quality traits found in studies were the following: weight (31), seed number (15) length (12) and diameter (10). Nonetheless, quality traits such as width (7), sugar content (5), commercial grade (5), firmness (3), circumference (3), color (2), shape (2), thickness (1), disease (1), germination (1), and minerals (1), were also found.

#### 3.2. Cucurbitaceae crops, pollinators identified and time periods found across studies

Out of the 37 studies, cucumber was the crop most studied (Fig. 2), followed by melon, watermelon, pumpkin and squash. Among pollinators, non-specific native bees were found in 47.5% of the studies, while honey bees in 34.4% and bumblebees in 9.8% of studies (Fig. 3). Other native bees were found in several studies (*Peponapis pruinosa, Nannotrigona testaceicornis tetragonisca angustula, Scaptotrigona depilis nannotrigona testaceicornis, Tetragonula iridipennis*), but amount to less than 5% of the pollinators found across studies. Most studies were performed during the 2000-2010 decade and the 2010-2020 decade, followed by the 1990-2000 decade, 2020-2021 and 1970s and 1980s (Fig. 4).

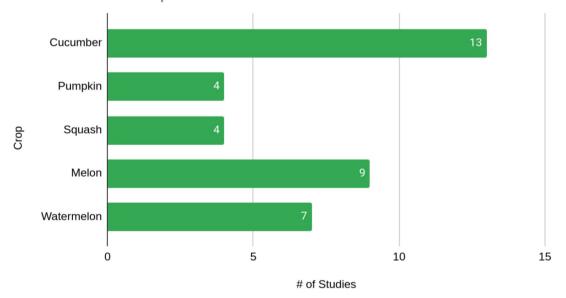




Fig. 2 - Species of Cucurbitaceae crops examined in the included studies.

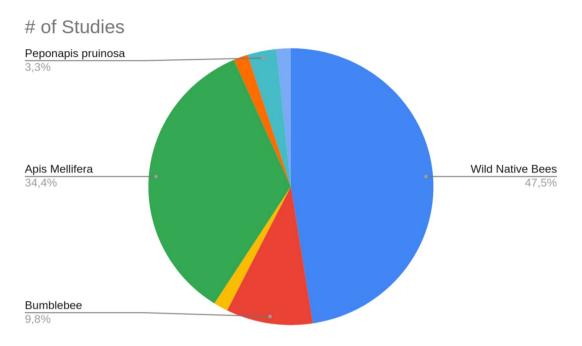
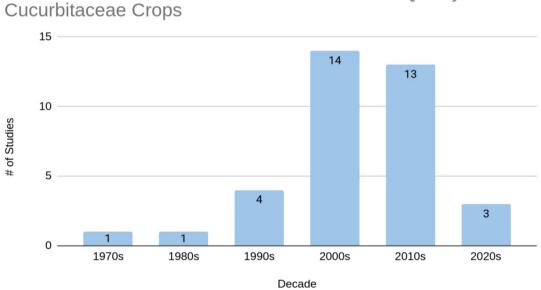


Fig. 3 - Percentage of pollinators investigated across the included pollination studies.



Number of Studies on Pollination Effect of Fruit Quality in

Fig. 4 - Distribution of included studies across time periods.

## 3.3. Importance of pollinators for Cucurbitaceae fruit quality traits

As shown in Fig. 5, the open pollination treatment versus the hand treatment and the exclusion treatment showed an improvement in the cumulative quality traits measured across studies. Fig. 6 shows the distribution of the number of studies and quality traits improved by open pollination versus exclusion. On the other hand, Fig. 7 shows the number of studies and quality traits where hand pollination had significantly improved fruit quality traits versus open pollination. Furthermore, Fig. 8 shows the number of studies where hand pollination improved quality traits over exclusion pollination treatment. Finally, Fig. 9 shows the comparison of all three pollination treatments' effect on fruit quality traits found across all studies.

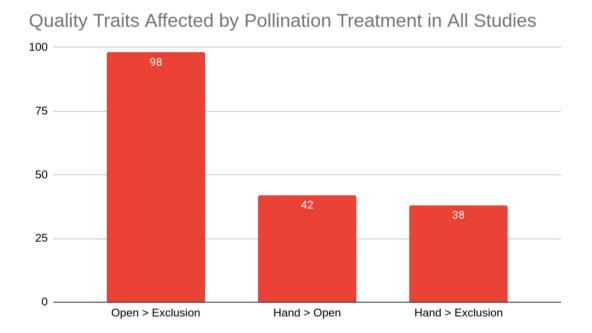
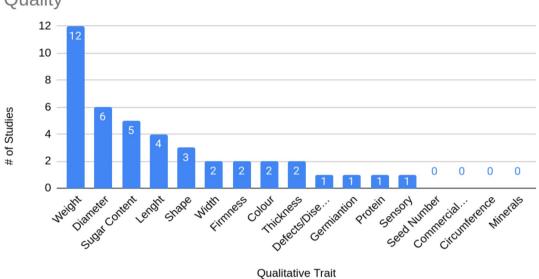


Fig. 5 - Number of trials with significant positive effect on Cucurbitaceae crops across all studies.



# of Studies where Hand > Open Pollination Improved Fruit Quality

Fig. 6 - Number of studies where open pollination treatment improved fruit quality traits over closed pollination.

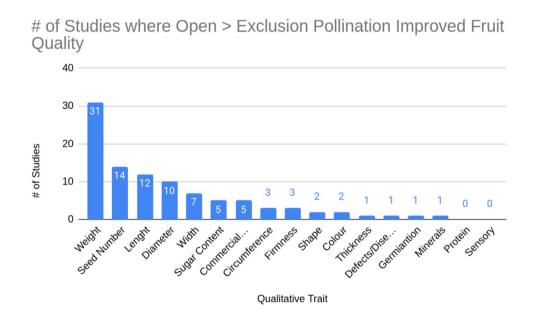


Fig. 7 - Number of studies where hand pollination treatment improved fruit quality traits over open pollination.



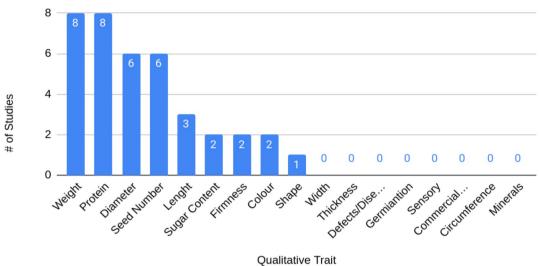
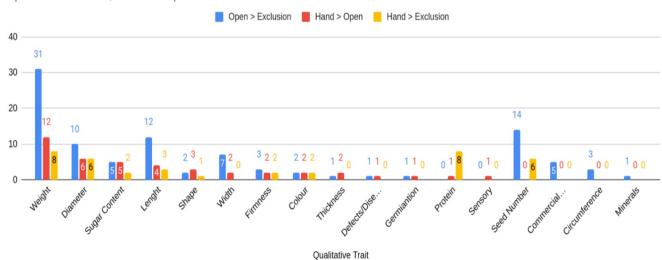


Fig. 8 - Number of studies where hand pollination treatment improved fruit quality traits over closed pollination.



Open > Exclusion, Hand > Open and Hand > Exclusion

Fig. 9 - Comparison of three pollination treatments and the qualitative fruit traits improved with respect to each other.

## 4. Discussion

#### 4.1. Changes in Cucurbitaceae crop research by region and time periods

During the course of this thesis, I analyzed how pollination affects the quality of Cucurbitceae crops. Overll, quality seems to better benefit from insect-mediated pollination than from wind (closed) pollination only, while hand pollination benefited fruit quality in certain cases with certain traits.

Even though the North American region had the most studies done on Cucurbitaceae crops (11), with the USA having the highest number of studies (9), the Asian continent has a more equal research distribution among its countries (Fig. 1). The increasing importance and population growth of several countries in Asia, explains the need for more research on crop pollination studies in order to ensure food security and sovereignty in the region. Furthermore, the Asian continent has a variety of landscapes and ecosystems, requiring vast research. Gaps in Cucurbitaceae crop pollination research in South America, Africa and Europe need to be filled.

An increase in Cucurbitaceae crop pollination studies over the last decades shows the importance and foresight of global food security issues in the coming decades due to climate change and a loss of biodiversity. Fig. 4 shows a substantial and continuous increase in research after the 2000s. As with this study, further research on pollination will take place in the coming decade.

## 4.2. Importance of pollinators for Cucurbitaceae fruit quality improvement

Overall, Fig. 6 and Fig. 9 show the quality traits improved by open pollination in comparison to closed pollination. Fig. 5 shows that 98 fruit quality traits improved with open > closed pollination in the 37 studies used in this literature review, compared to 38 quality traits improved by hand > closed pollination.

Weight was by far the quality trait most commonly shown to improve by open pollination. Seed number was also shown to significantly improve in open pollination, even compared to hand pollination. Certainly, a larger number of seeds might not necessarily mean better fruit quality, but it can provide with more abundant and diverse genetic material for next season's sowing. Overall fruit size (length, width, diameter), was greater in open pollination compared to hand or closed pollination. These traits are the most valued quality component for market and commercialization.

Hand pollination did provide quality trait improvement in three main categories: weight, diameter, and sugar content, above open or closed pollination. Protein content was the single quality trait that hand pollination exceeded above open or closed pollination. One study in Thailand found that cucumber fruit

set was higher in hand pollination than open pollination, however, other traits such as weight, diameter, and length were significantly higher in open pollination (Sawatthum et al. 2017).

Another study in Sub-saharan Africa found that "extra [hand] pollination significantly increased the probability of a plant producing a second fruit of a size the farmer could sell at the market, and also the fruit sugar content" was higher than in the open pollination treatment (Sawe et al. 2019). The authors conclude that pollination abundance might be poor in their ecosystem.

Other studies used novel pollination methods to test its efficacy. One study in particular evaluated the effect 1) CPPU solution pollinator on ovaries, 2) artificial pollination by hand and 3) honeybee pollination had on melon fruit sugar and amino acid content (Huang et al. 2017). The Chinese researchers concluded that honeybee pollination produced tastier and heavier fruits and with a higher sugar content than the other two treatments.

One Polish study (Gajc-Wolska et al. 2011) found better quality traits (firmness, diameter and minerals) in cucumber cultures with bumblebee pollination compared to open pollination.

Another study used honeybees in comparison with other pollination treatments. In India Thakur & Rana (2007) had three pollination treatments for measuring cucumber quality traits: open pollination, hand pollination and honeybee pollination. In terms of misshapen fruits, open pollination gave the highest number (20%) followed by hand (14%), followed by honeybee pollination (8%). Furthermore, honeybee pollination gave heavier fruits, bigger fruits and with more seeds, compared to the other treatments.

#### 4.3. Diversity of pollinators

As mentioned in before, Cucurbitaceae crops are highly dependent on pollinators for successful fruit production since they have a male and female flower on each plant. In these studies, closed/exclusion treatment meant that the pollination either happened through selfing or wind factors. Even though honey bees (*Apis Mellifera*) are one of the most common and efficient types of pollinators (Cane et al., 2011, Free 1993), they are found in only one-third of the studies, while bumblebees account for less than 10% of the cases.

In Turkey, a study by Dasgan et al., (1999), found that the weight, height, diameter, and number of seeds of melon crops were significantly higher in bumblebee pollination than honey bee pollination under protected cultivation. A study in New Zealand by Fisher & Pomeroy (1989) also found that Bumblebee pollination alone could produce expotable muskmelons due to their quality weight.

A study in Brazil (Dos Santos et al., 2008) using native stingless bees *Scaptotrigona aff. Depilis Moure* and *Nannotrigona testaceicornis* Lepeletier, found cucumber production under greenhouses to have less imperfections and diseases than the open field pollination treatment. However, a study (Nicodemo et al., 2013) with parthenocarpic cucumbers pollinated in greenhouses with stingless bees Jataí (*Tetragonisca angustula*) and Iraí (*Nannotrigona testaceicornis*) and Africanized honey bees, found no significant difference in in weight between bees, but there are differences in length and diameter.

Another study using stingless bee *Tetragonula iridipennis* in southern India (Tej et al. 2017) found that cucumber production under greenhouse conditions had significantly higher weight, girth, and length, compared to greenhouses without stingless bee colonies.

Furthermore, a study in Greece (Garantonakis et al. 2016) on watermelon production, found that even though native bees *Lasioglossum* spend three times long on each flower compared to *Apis Mellifera*, fruit quality traits of mean weight, brix, number of seeds per fruit and weight of seed, did not differ between both pollination treatments. This further strengthens the hypothesis that native bee populations can sustain commercial production of highly dependent pollination crops such as watermelon.

Finally, a study in Israel (Sadeh et al. 2007) using native bees in greenhouses for melon pollination, found that even though "visit durations per flower were shorter for *X. pubescens* than for honeybees, pollination by both bees resulted in similar fruit mass and seed numbers. However, *X. pubescens* pollination increased fruit set threefold as compared to honeybee pollination.

Over half of the studies used wild native bees as pollinators, primarily due to the open field pollination treatments. However, what is important to highlight is that, as we approach an ecologically unstable world, we need to focus on local solutions for ensuring food security, which means local biodiversity as a first priority. The conservation and nurturing of native pollinators is of the utmost importance. This can be achieved through landscape management, agro-chemical management and integrated pest management (IPM).

## 5. Conclusions

Pollinators are key agents in food production as well as fruit set and quality traits for Cucurbitaceae crops. In most cases, open pollination with native pollinators was sufficient for marketable fruit quality traits. However, in certain regions where pollinators are scarce, like in Sub-Saharan Africa, an immediate alternative could be hand pollination or bee hive implementation in protected cultures. This has to go through a cost-benefit analysis before implementing artificial or costing pollination services.

Again, the problems and solutions are local, no single receipt can be imposed on every agroecological system with its varying social and economic contexts. Nonetheless, several studies mentioned in the introduction, have shown that an adequate management of landscape habitats can enhance pollination abundance and diversity (Pardo et al. 2020, Woodcock et al. 2019)

Successful and commercially adapted Cucurbitaceae fruit pollination is effective with either honeybees or native pollinators. Bumblebees also prove to be effective pollinators for Cucurbitaceae crops. Future research should focus on other agricultural factors such as habitat management for pollinators, agrochemical use and a deeper understanding on local pollinator diversity under climate change pressures.

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