Diagnostic Survey of Honeybee Diseases, Pests and Predators in Bale Zone Southeast Oromia Region, Ethiopia

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Abstract

A cross-sectional study was conducted on parasitic honeybee diseases, pests and predators in Bale. For the study questionnaires and diagnostic survey were employed. A sample of adult worker bees and brood for major honeybee diseases were taken and analyzed in veterinary laboratory of respective districts of the study. SPSS version 20 was used for data analysis using descriptive statistics (frequency, percentage and chi-square. From the present study, the major challenge declared by beekeepers includes pests and predators, lack of bee forage, absconding and mass death of bee colonies. Regarding to honeybee pests and predators, the most important identified were honey badger, spiders ant, wax moth, bee-eater birds, small hive beetles and monkeys. The study indicated the overall mean prevalence of Nosema apis, Amoeba mellifera, Varro mites, bee lice, wax moth, small hives beetles were 79.17 %, 79.16%, 72.22%, 18.06%, 40.27% and 54.2 % respectively. However, the current diagnosis was not detected AFB, EFB, and trachea mites during laboratory sample analysis. Agro ecology and hive types were identified as risk factors for prevalence of honeybee diseases and pests. Further study on prevalence of seasonal honeybee diseases and outbreak of honeybee diseases, and pests is could be very important. **Keywords:** Honeybee, Nosema apis, Malpighamoeba mellificae, diagnostic, disease, pests

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Introduction

Honeybees are essential pollinators of wild and cultivated plants and are vital to food production and biodiversity conservations. Honeybees provide a vast range of products for humans from honey to other bee products, such as pollen, beeswax, royal jelly, venom, etc. *A. mellifera* are commercially valuable as essential plant pollinators, and for the high demand for their products, like honey and wax. Honeybee colonies existing in the wild away from man's control produce a small surplus of honey above their requirements signifying beekeeping is much more productive and profitable if they only managed properly (Moeller, 1982).

Like other all living organisms, honeybees can be infected and infested with harmful diseases and pests respectively. The decline of the honeybee population due to agricultural chemicals, pests, predators, and diseases is of great concern to many countries around the world, including those in Asia (Kajobe *et al.*, 2016; Abdulhay & Yonius, 2020). According to Kajobe *et al.* (2016) and Abdulhay and Yonius (2020) in tropical regions, beekeeping is threatened by various pests, predators, and diseases, which often lead to economic losses. Most of the time pests, predators, and diseases interact with each other and affect the health, performance of the honeybee colonies, and reduces the yield of bee products (Forfeit *et al.*, 2015). Hence, it is vital to maintain a healthy honeybee population to ensure the supply of honey and other bee products to adequately meet the domestic and international market demand. The recent high honeybee colony losses in many parts of the world are associated to the vulnerability of honeybees to parasitic mites, fungi, viruses, and bacteria (Bradbury, 2009). These pathogens and parasites can have harmful effects on honeybee health and the services they offer, which in turn can lead to severe economic losses (Genersch, 2010).

Even though, the majority of pathogens and parasites affecting honeybees have almost worldwide distribution (Ellis and Munn, 2005). The most commonly known honeybee diseases reported to exist in Ethiopia are Nosema *apis*, amoeba (Gezahegn T and Amssalu B, 1991) and chalk brood (Amssalu and Desalegn, 2005).

Although research on diagnosing honeybee diseases and pests was done in Ethiopia at various times by various researchers, there were several areas that had not yet been well covered. Bale zone of Oromia regional State is noteworthy among the areas that have not yet been thoroughly researched since it has a significant potential for beekeeping. To fully take advantage of the beekeeping resources available in the Bale zone, it is prudent to conduct honeybee disease and pest diagnostic studies in this area. The goal of the current study was to identify the prevalent diseases, predators, and pests in the Bale zone as well as the risk factors that go along with them.

Materials and methods

Description of the study areas

The study was conducted in Bale zone Southeast Oromia Regional state, Ethiopia. Honeybee pathogens and pests were diagnosed and assessed from 2020-2021 in three selected districts namely Dinsho, Dello-Menna, and Goro

districts.

A cross-sectional study was carried out from 2020 to 2021 in three selected districts in Bale Zone to identify the major honeybee diseases, Pests and predators through inspection and examination of collecting samples from the colonies. Questionnaire survey was carried out during diagnostic study to determine honey production system and constraint caused due to pests and predators. Types of hive and agro-ecology were considered as explanatory variables (risk factors), and tested whether they have an impact on occurrence of honey bee disease and pests. Honeybee hive was categorized as Traditional, Transitional and Modern hives. Three altitude categories were considered: highland (>2400 meters), Midland (1800 to 2400 meters) and lowland (lowland (1800 meters) above sea level). Finally, prevalence for apiary and colony levels was calculated following the protocols of Vanenglesdorp *et al.* (2013):

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Sampling method and sample size determination

For this study, multi sampling techniques were employed to collect sample of honeybee diseases and pests from suspected bee colonies in the study areas. Districts were selected using purposive sampling method based on their potential of beekeeping. Three rural kebeles from each district Abakera, Mio and Zallo Ababa from Dinsho district; Gomgoma, Cirri, and Erba from Delo Menna district and Balle Gadulla, Balle Anole and Dayu Abergada from Goro district were also selected purposively based on their beekeeping potential and representativeness of highland, midland and lowland agro-ecologies. 3 to 4 honeybee colonies were selected randomly from each apiary and inspected for pests and diseases at field. Brood and adult bees were also taken from the same colonies. In general 72 honey bee colonies were diagnoses in this study.

Sample Collection and Questionnaire survey

In this study, a semi structured questionnaire was prepared and administered to collect information from the beekeepers. Both primary and secondary sources of data were used. Secondary data were obtained from reports of zonal and districts livestock and fishery resource development Office, and other published and unpublished materials. Primary data was collected using questionnaire survey. Information about trends of beekeepers using a structured questionnaire.

Diagnosis for major honeybee diseases

Diagnosis of major honeybee diseases was conducted at field through inspection and examination of samples at laboratory of perspective districts Veterinary for Chalk brood, American foul brood, European foul brood, Nosema, Amoeba using their respective protocols.

Diagnosis for Chalk Brood (CBD)

Both external and internal inspection was conducted for the presence of chalk brood clinical symptoms. A dry scale with white to dark color molds and chalk brood mummies was carefully inspected in the comb cells and on the bottom boards of the hives. From each suspected colonies samples were collected from the comb cells and brought to lab for examination. The suspected samples placed on the slid and then moistened with a drop of distilled water. The suspension was examined for spores and/or, spore ball and/or spore cysts of *Ascosphaera apis* using light microscope under magnification power of 40 x.

American Foul Brood (AFB) and European Foul brood (EFB)

For AFB and EFB diseases in selected apiaries 3 to 4 colonies were inspected internally for the major clinical symptoms. Typical clinical symptoms like irregular brood arrangement, sunken and dark capping with puncture holes, dead and decayed larvae with dark "scales" and slight odor suspected was examined for the occurrence of AFB. Similarly, twisted larvae with creamy-white guts visible through the body wall, melted and yellow white larvae with unpleasant sour odor and loosely-attached brown scales were directly observed for the infected colonies of EFB. Furthermore, match stick test (stretch test) were employed to observe the robby thread stretching for the typical clinical symptoms of bacterial diseases. For the suspected brood showing one of the above important clinical symptoms, brood samples was prepared on microscopic slide for further laboratory diagnosis according to Primefact, 2009 (www.dpi.nsw.gov.au/primefacts.The samples was examined under microscope for the presence of *Paenibacillus larvae* and *Melissoccocus pluton* in positive samples AFB and EFB, respectively using Zeiss AxioVert A.1 light microscope under oil immersion (magnification power of 100X).

Protozoa diseases (Nosema apis and Amoeba)

For suspected colonies, a sample of 30-60 worker adult honeybees was collected from the hive entrance following

Fries *et al.* (2013) procedure. The abdomens were placed and grounded by mortar containing 5-10 ml distilled water until an even suspension is formed using pestle. The mortar and pestle were thoroughly cleaned before being used again. A loop of suspension were placed on microscopic slid using the sterilized loop and covered with cover slid. The suspension was examined under microscope using 40-magnification power for the presence of Nosema slippery and rod shaped spores and for Amoeba round cysts and spore balls.

Diagnosis for honeybee Ecto-parasites

Examination for Varroa mites

For diagnosis about 200-300 adult workers bees samples were collected from suspected bee colonies. The samples were examined using shaking method by adding detergent solution (10 ml of detergent is used to 1000ml of water solution) was poured into each of jar containing bees up to half of the jar get full. Then shaking for one minute until the varroa mites dislodged from adult honeybees. Straining the solution through a ladle (8-12 mesh) to remove the bees and then sieving the solution through tea strainer to collect varroa mites. Finally, the presence of varroa mites was checked either by necked eye or by using hand lens count according to Cramp, 2008.

For examination of varroa mites in bee brood, samples were taken from the suspected 3-5 bee colonies of brood comb 5cm x 5cm size. About 100 pupae was removed from their cells using forceps or match stick test and checked for the presence of varroa mites. At the end, number of varroa mites per diagnosis samples was recorded and determined the infestation level per colony.

Bee lice

For the examination of bee lice, the same procedure with varroa mite was followed.

Examination for Tracheal mite

For diagnosis of tracheal mites, an average samples of 60 worker bees' were collected from suspected honeybee bee colonies in order to detect 5% of diseased bees with 95% confidence (Fries I, 1988). The abdomen of bees was held between thumb & forefinger & push off the head with forelegs, cut parallel a small disc of the breast put all the discs in small bottle containing 10% Potassium and boil in water bath for 4-8 minutes put the discs on wire gauze. Then the discs was rinsed in tape water to clean all muscles dissolved, put on slide with few drops The trachea suspension was examined for infested part and *Acrapis woodi* under to examine through microscope using magnification of 40X

Diagnosis of major honeybee pests

The occurrence of major honeybee pests in the all the study areas was determined through internal and external hive inspections. In addition, for clinical symptoms and infested combs, adult and larvae of small hive beetles and wax moth was detected in the hive through inspection of the beehives described by Neumann *et al.* (2013). Infestation and prevalence

Infestation (%) =
$$\frac{Positive number of bee colonies}{The total number of bee colonies examined} X100$$

Prevalence (%) = $\frac{Number of sites diseases positive}{The total number of sites surveyed} X100$

Ranking of the different types of beekeeping constraints obtained in the study were done by using the rank index formula as described by (Musa *et al.*, 2014).

Data Management and Statistical Analysis

The collected data were entered & stored into Microsoft Excel program for further analysis. SPSS version 20 statistical package was used to analyze the data. Summarized data was presented in the form of tables and figures. For the survey data, analysis was done by using descriptive statistics and the rank index formula as described by (Musa *et al.*, 2014). Chi-square test was used to assess the association of the risk factors with the prevalence of the disease and pests. Statistical significance was set at $p \le 0.05$ with 95 % CI

Result and Discussion

Socio-economic characteristics of the Respondents

Out of the total sample respondent about 96.3% were male headed household and only 3.7% were female headed household. This indicated that most of beekeeping in study area is male's job and it is probably because of traditional taboos that females are not allowed to carry beekeeping activities in the study areas. A similar issue was reported by Tesfaye *et al.* (2017) from the high lands of Bale and Workineh (2006) also reported beekeeping as male-headed households dominated activity in the Northern part of Ethiopia.

According to the current findings, beekeepers' ages range from 30 to 80 years old, with a mean age of 49.17 years (Table 1). This result demonstrated that beekeeping activities were carried out by different age groups and

more actively by younger age groups. This result in line with the report of Tesfaye *et al.* (2017) and Chala *et al.* (2012) whose reported from high land of Bale and Jimma Zones respectively. Table1. Age of sample respondents in the study areas

| Districts | Total sample size (N=81) | | | |
|--------------|--------------------------|---------|---------------|--|
| Districts | Maximum | Minimum | Mean \pm SE | |
| Dinsho | 80.00 | 32.00 | 52.00±3.29 | |
| Goro | 70.00 | 30.00 | 44.71±3.27 | |
| Dello Menna | 72.00 | 30.00 | 50.80±3.18 | |
| Overall mean | 74.00 | 3.67 | 49.17 ±3.25 | |

Source: Survey result, 2022

Source of honeybee colonies and apiary site place

The strategies used by beekeepers to start their beekeeping businesses varied depending on their economic capacity and agro-ecologies where they live. As beekeepers reported about 82.72% of them began their beekeeping activity by catching swarms, while 7.4% of the sample respondents started by getting gift from their parents and 9.88% of respondents started by getting colonies gift from their parents and caught their own swarm. This indicated that most of the respondents engaged in beekeeping by getting colonies from their locality. This result showed that swarm catching was the main source of colonies in the study area. Similarly, Tesfaye *et al.* (2017) and Chala *et al.* (2012) reports that beekeepers started beekeeping in Bale and Jimma Zones by swarm capturing 98.3% and 87.8% respectively. The majority of respondents (92.6%) stated that they were started beekeeping as sideline activities, whereas only 7.4% of beekeepers used beekeeping as their primary source of income. This finding demonstrated that beekeeping can typically integrate with other farming activities.

Of all the responders about 39.51% keep their colonies in their backyards, whereas 24.93% keep in forest by hanging on long trees, 16.05% in apiary sites, 18.52% of beekeepers install their honeybee colonies in their backyards or hang on trees in forest (Table 2). Earlier research found that 50% of beekeepers in the Bale zone set up honeybee colonies in their backyards (Tesfaye *et al.*, 2017). Many scholars reported that the majority of beekeepers in various regions of Ethiopia set up their honeybee colonies in their backyard (Tessega, 2009; Gidey *et al.*, 2012; Nebiyu and Messele, 2013 and Niguse, 2015). Table 2. Apiary sites location

| Diaga hashaaning | Total sample size (N=81) | | |
|-------------------------------|--------------------------|-------------|--|
| Place beekeeping | Frequency | Percentages | |
| Backyard | 32 | 39.50 | |
| hanging on trees in forest | 21 | 25.93 | |
| Backyard and hanging on trees | 15 | 18.52 | |
| Apiary sites | 13 | 16.05 | |
| Total | 81 | 100.01 | |

Source: Survey result, 2022

Trends of beekeeping in the study areas

As the sample respondent mentioned about 69.14% beekeeping trend was decline in the number of honeybee colonies, while 19.75% of respondent said that the population of honeybee colonies had not changed and only 11.11% of the respondents stated that honeybee colony populations were and upward trending. In addition, about 74.07% of the respondents claimed that honey yield was decline, while 16.05% said there were no changes in honey production and the remaining 9.89% said that honey yield in the research areas was increasing (Table 3). This result is agreement with report of Tesfaye *et al.* (2017) who reported that honey bee population and honey yield were decreasing from time to time in the Bale Zone. Table 3. Trends of honeybee colonies in the study areas

| | Total sample size (N=81) | | | | | |
|----------------------|--------------------------|-------------|-------------|-------------|--|--|
| Trends of categories | Honey | bees colony | Honey yield | | | |
| | Frequency | Percent (%) | Frequency | Percent (%) | | |
| Decreasing | 56 | 69.14 | 60 | 74.07 | | |
| Increasing | 9 | 11.11 | 8 | 9.89 | | |
| Unchanged | 16 | 19.75 | 13 | 16.05 | | |
| Total | 81 | 100.00 | 81 | 100.00 | | |

Source: Survey result, 2022

The respondent reason out that many factors are responsible for the declining of honeybee production and they have mentioned that caused by climate change (drought, shortage of feeds, lack of water), unwise use of agrochemical, pests and predators and diseases the detail reason presented in table 4. This result agrees with reports by Shunkute *et al.* (2012), Haftu and Gezu (2014) and Tesfaye *et al.* (2017) in Southern Nation and Nationalities People (Keffa, Sheka and Bench-Maji Zones), Tigray region and Bale zone southeast Oromia region respectively. Table4. Major causes of honeybee colonies and honey yield declining

| r wore in major ewweep or money oue coronnes w | | | | | 2 | | | | |
|--|------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|------------|------|
| | 1 st | 2 nd | 3 rd | 4 th | 5 th | 6 th | 7 th | Rank index | Rank |
| Pests and predators | 18 | 54 | 72 | 54 | 0 | 126 | 90 | 0.698 | 1 |
| Lack of bee forage | 84 | 56 | 14 | 56 | 28 | 84 | 56 | 0.519 | 2 |
| Absconding | 18 | 24 | 30 | 6 | 24 | 60 | 6 | 0.265 | 3 |
| mass death of bee colonies | 55 | 11 | 22 | 11 | 33 | 11 | 55 | 0.252 | 4 |
| Unwise use of agro-chemicals (Pesticides) | 36 | 18 | 45 | 18 | 0 | 36 | 18 | 0.238 | 5 |
| Diseases | 26 | 26 | 26 | 0 | 39 | 39 | 0 | 0.229 | 6 |
| Drought | 16 | 40 | 32 | 24 | 0 | 16 | 0 | 0.198 | 7 |
| | | | | | | | | | |

Source: Survey result, 2022

Index = sum of (7*ranked 1st+ 6* ranked 2nd+5* ranked 3rd+4* ranked 4th+3* ranked 5 th+2* ranked 6th+1* ranked 7th) for individual and predators divided by the sum of (7*ranked 1 st+ 6* ranked 2nd+5* ranked 3rd+4* ranked 4th+3* ranked 5th+2*ranked 6th+1* ranked 7 th) for over all Constraints

Categories of Honeybees Behaviour in Study areas

According to the response of sample respondents, honeybees behaviour in the study areas were categorized as 25.93% aggressive, 18.52% medium, 11.11% docile, 25.93% medium and aggressive, 11.10 docile and aggressive, 7.41% docile, medium and aggressive. Similarly, Dubale BT (2017) reported that honeybee's behaviour as very aggressive (27.2%), aggressive (51.1%) and docile (21.7%) and depending on their stinging behavior. Table 5. Honeybees' behaviour in the study areas

| Total sample si | ize (N=81) | |
|-----------------|--|--|
| Frequency | Percentage (%) | |
| 21 | 25.93 | |
| 21 | 25.93 | |
| 10 | 18.52 | |
| 15 | 11.11 | |
| 15 | 11.10 | |
| 6 | 7.41 | |
| 81 | 100.00 | |
| | Frequency 21 21 10 15 15 6 | $\begin{array}{cccccccccccccccccccccccccccccccccccc$ |

Source: Survey result, 2022

Inspection of Honeybee colonies

Sample respondents were requested to describe the frequency of inspection of their apiaries and colony status, and the majority of respondents (83.3%) were inspected their apiary sites, while 16.7% did not. It was mentioned that both internal and external hive inspections were practiced by beekeepers in the study areas. However, the majority of beekeepers in the study areas inspected their honeybee colonies externally (72.22%), internally (18.52%) and both internal and external inspections (9.26%) with frequencies of 51.89%, 33.33% and 14.85% some times, frequently and rarely respectively. This result in line with the findings of Tessega (2009)who did the study in Burie district of Amhara.

Major Honeybees Pests and Predators

Presence of pests and predators were reported by sample respondents in the study areas. The major honeybee pets and predators mentioned were Honey badgers (*Mellivora capensis*), Spiders (*Arachnids*), Ants (*Dorylus fulvus*), Wax moths (*Acheroea grisella*), Bee-eater birds (*Meropidae*), small hive beetles (*Aethina tumida*), and Monkeys. When they were requested to rank these pests and predators according to their economic importance ranked as in table 7. According to the ranking index honey badgers were took the most problematic and dangerous predator of honeybees and followed by spiders (Table 6) whereas monkey has the least impact on honeybees. Different authors have reported similar ideas (Shunkute *et al.*, 2012; Chala *et al.*, 2012; Tesfaye *et al.*, 2017).

| Table 6 | The major | nests and | nredators | of honeyhees | in the study areas |
|----------|-----------|-----------|-----------|--------------|--------------------|
| Table 0. | The major | pests and | predators | of noneybees | in the study areas |

| Major pets and predators | Rela | Relative level of importance | | | | | | Index | Ranks |
|------------------------------------|-----------------|------------------------------|-----------------|-----------------|-----------------|-----------------|-----------------|-------|-----------------|
| | 1 st | 2 nd | 3 rd | 4 th | 5 th | 6 th | 7 th | | |
| Honey badger (Mellivora capensis) | 98 | 60 | 45 | 28 | 18 | 10 | 3 | 0.693 | 1 st |
| Spiders (Arachnids) | 91 | 60 | 35 | 40 | 15 | 12 | 3 | 0.677 | 2 nd |
| Ant(Dorylus fulvus) | 77 | 54 | 60 | 24 | 24 | 12 | 2 | 0.669 | 3 rd |
| Wax moth (Acheroea grisella) | 91 | 60 | 40 | 24 | 18 | 14 | 4 | 0.664 | 4 th |
| Bee-eater birds (Meropidae) | 63 | 42 | 65 | 32 | 21 | 10 | 5 | 0.630 | 5 th |
| Small hive beetles(Aethina tumida) | 56 | 30 | 50 | 40 | 27 | 16 | 4 | 0.590 | 6 th |
| Monkey | 49 | 36 | 40 | 36 | 15 | 16 | 11 | 0.537 | 7 th |

Index = sum of (7*ranked 1st+ 6* ranked 2nd+5* ranked 3rd+4* ranked 4th+3* ranked 5 th+2* ranked 6th+1* ranked 7th) for individual and predators divided by the sum of (7*ranked 1 st+ 6* ranked 2nd+5* ranked 3rd+4* ranked 4th+3* ranked 5th+2*ranked 6th+1* ranked 7 th) for over all Constraints

Traditional strategies for controlling of honeybee pests and predators

During study period, several techniques were stated by sample respondents to control pests and predators in the study locations. When they were asked how to control pests and predators in your area, the respondents were mentioned using ash around hives stands for the most common pests, attaching smooth iron sheets to the trunks of trees where hives are hanging, hanging hives on long trees with very smooth bark so that honey badgers cannot climb them, using dogs for back yard, and killing honey badgers by using *wotmed* (Table 7). Different researchers have reported that beekeepers in different part of the country were used several defense strategies to keep their honeybees safe from pests and predators (Chala *et al.*, 2012; Dabessa and Belay, 2015; Tesfaye *et al.*, 2017) Table 7.Traditional practices of controlling major honeybee pests and predators by beekeepers

| Pests and predators | Traditional controlling mechanisms /practices |
|---------------------|---|
| Honey badger | Use of dog for chasing, use of "wotmed" to kill, fencing the apiary site with strong |
| | fence, hanging hives by rope on long trees |
| Spiders | Cleaning apiary site always, removing the spider webs, putting ash under the hive stand |
| Ants | Applying ash under hive stand, cleaning apiary site |
| Wax moth | Supply supplementary feeding and water to the colonies to be strong, fumigating the |
| | hive, removing the old comb from hives, and cleaning beehives |
| Birds | Frightening birds from the area, putting like tallow, mastic, and plastic on hive |
| | entrance, placing wheat seed or barely, putting an image of a human near the hives |
| | using cloth |
| Small hive beetles | strengthening the colony or keeping strong colonies, removing weak colonies, cleaning |
| | apiary, smoking/fumigating the hive |
| Monkey | Hanging beehives on a branch of long trees by ropes, keeping beehives near home |
| | steady |
| Bee lice | Fumigate the hive with materials like tobacco, dung and grass, making colonies strong, |
| | supplying additional food for weak colonies |

Source: Survey result, 2022

Laboratory Diagnosis of Honeybee Diseases and Pests

Honeybee diseases

Prevalence of Nosema apis and its associated risk factors

The present study revealed that the overall prevalence of *Nosema apis* in Bale Zone was 79.17% and ranges from 68.18% to 86.96% (Table 9). There was statistically significant different (p<0.05) between agro-ecologies. The highest prevalence of *Nosema apis* was observed 86.96% highland followed by 81.48% in midland and the least was observed 68.18% in lowland. The result showed that honeybee colonies infected by *Nosema apis* was found healthy and active in their production performance. The same result was reported by Semere *et al.* (2021) in Kaffa zone. The current finding is also in agreement with the finding of (Amssalu , 2012; Nega *et al.*, 2019) who stated that increase in humidity and rainfall limit honeybees to fly out for cleansing, which in turn enhances spread of the disease among the members and autoinfection.

There was a significant difference on prevalence of *N.apis* honeybees colonies kept in different behive types. The highest prevalence was observed 85.71% in modern behives, followed by 72.22% traditional behives while the lowest was found 66.67% (Table 10) transitional behives during the study period. This variation might be related to various operations on behives management practices; exchange of bee equipment, placement of behives may conducted by beekeepers. Desalegn and Kebede (2005) stated that *Nosema apis* disease was more prevalent in the modem behives (72.2%) than in traditional behives (41.3%) and transitional behives (35.3%)

systems. Similarly, Amsalu (2012) reported that *N. Apis* infection rate in the central Ethiopian highlands had reached up to 82%.

| Variables | Category | No. of sampled colonies | Prevalence (%) | x ² | p-values |
|---------------------------|---------------|-------------------------|----------------|-----------------------|----------|
| Beehive types | Movable frame | 42 | 36(85.71%) | | |
| | Transitional | 12 | 8(66.67%) | 2.76 | 0.252 |
| | Traditional | 18 | 13(72.22%) | | |
| Agro-ecology | Highland | 23 | 20(86.96%) | | |
| | Midland | 27 | 22(81.48%) | 2.54 | 0.28 |
| | Lowland | 22 | 15(68.18%) | | |
| Overall prevalence | | 72 | 57(79.17) | | |

Table 9: Prevalence of *Nosema apis* and Associated risk factors in the study areas

Source: Survey result, 2022

Prevalence of Malpighamoeba mellificae and Associated Risk Factors

In the current study, the overall prevalence of amoeba (*M. mellificae*) disease was 79.16% (Table10). The result indicated that amoeba (*M. mellificae*) disease was higher in movable frame hives (83.33%) followed by transitional hives (75.00%) and traditional hives (72.22%). Furthermore, there was no a statistically significant difference between these hives (p > 0.578). This might be due to several in management practices like exchange of old combs easy operation bee equipment and use of traditional systems. The current result agrees with the finding of Begna and Kebede (2005) who reported the prevalence of amoeba was high in the modern beehives (88.9%) than in the traditional (61.9%) and transitional (47.1%) system.

The present investigation had shown that the prevalence of Amoeba was higher in the highland (82.61 %) than midland land (77.78%) and lowland (77.27%) agro-ecologies (Table 10). There was no a significant difference on the prevalence of Amoeba disease among agro-ecologies (p > 0.05). Amssalu (2012) stated that Amoeba diseases was widely distributed and identified in the most places of the country throughout the year. This result was in line with the previous studies reported in different part of the country for prevalence of Amoeba 88 % Oromia region, 95 % Amhara region and 60 % Benishangul-Gumuz region (Yohannes A, 2009). Table10: Prevalence of *Malpighamoeba mellificae* and associated risk factors

| Variables | Category | No. of sampled colonies | Prevalence (%) | df | x ² | p-values |
|---------------------|--------------|-------------------------|----------------|----|-----------------------|----------|
| Beehive types | Movable fram | e 42 | 35(83.33%) | | | |
| ••• | Transitional | 12 | 9(75.00 %) | 2 | 1.095 | 0.578 |
| | Traditional | 18 | 13(72.22%) | | | |
| Over all prevalence | e | 72 | 52(79.16) | | | |
| Agro-ecology | Highland | 23 | 19(82.61%) | | | |
| | Midland | 27 | 21(77.78%) | 2 | 0.245 | 0.885 |
| | Lowland | 22 | 17(77.28%) | | | |
| Over all prevalence | e | 72 | 52(79.16 %) | | | |

Source: Survey result, 2022

Prevalence of honeybee pests

Prevalence of varroa mites and associated risk factors

The study result showed that the prevalence of *varroa* mites was observed 95.45 % lowland followed by 66.67 % midland while the lowest was found 56.2 % highland (Table 11). The prevalence of varroa mite in Bale zone ranges from 56.52% to 95.45% with an average of 72.22%. Moreover, there was statistically significance difference (p<0.05) among agro-ecologies. This indicated that hot environment was very conducive for the spread of varroa mite's populations. This result is higher than the earlier research finding of 82% varroa mite prevalence in the Tigray Region Begna (2015). Thus with this prevalence rate, varroa mite is less likely to impact honeybee health and its honey production in the study areas.

The present result revealed that the prevalence of *varroa* mites was higher in transitional behives (83.33 %), than modern behives (73.81 %) and traditional behives (61.11 %) (Table 11). The prevalence of varroa mites high in transitional and modern behives might be due to transmission of the pest during exchange of bee equipment's and management of bee colonies like feeding. Similarly, Tsegaye (2015) noted 94.2%, 84.8%, and 79.85% in movable frame hives, intermediate hives, and traditional hives, respectively, in the eastern regions of the Amhara Region.

| Variables | Category | No. of sampled colonies | Prevalence (%) | df | x ² | p-values |
|---------------------|---------------|-------------------------|----------------|----|-----------------------|----------|
| | Movable frame | 42 | 31 (73.81%) | | | |
| Beehives types | Transitional | 12 | 10(83.33%) | 2 | 1.899 | 0.387 |
| V 1 | Traditional | 18 | 11(61.11%) | | | |
| Over all prevalence | | 72 | 52(72.22 | | | |
| Agro-ecology | Highland | 23 | 13 (56.52 %) | | | |
| | Midland | 27 | 18(66.67%) | 2 | 9.16 | 0.01 |
| | Lowland | 22 | 21(95.45%) | | | |
| Over all prevalen | ce | 72 | 52(72.22 %) | | | |

Table 11. Prevalence of Adult varroa mites and associated risk factor

Source: Survey result, 2022

Prevalence of varroa mites on Brood and associated risk factor

The current result showed that the overall prevalence of varroa mites in brood was 18.06%. The highest varroa mites in brood was observed 27.78% in traditional behives and followed by 25.00% in transitional behives and 11.90% in modern behives. Guesh (2015) reported that traditional hives had a higher frequency of varroa mites than movable frame hives.

Concerning agro-ecology, the highest vorroa mite's population in brood was found in midland (33.30) and followed by lowland (9.09%) and the least populations found in high land (8.69%) agro-ecology (Table 12). The present finding showed varroa mites found in all agro-ecologies throughout the year and higher during brood rearing season. Begna *et al.* (2016) stated that the population of varroa mites recovered was low during the dry season and low brooding tendency and growth of brood population mites depend on honeybee brood production. Table12. Prevalence of varroa mites in brood and associated risk factor

| Variables | Category | No. of sampled colonies | Prevalence (%) | df | \mathbf{x}^2 | p-values |
|--------------------------|---------------|-------------------------|----------------|----|----------------|----------|
| | Movable frame | 42 | 5(11.90%) | | | |
| Beehives types | Transitional | 12 | 3(25.00%) | 2 | 2.615 | 0.270 |
| ••• | Traditional | 18 | 5(27.78%) | | | |
| Over all prevalence | ce | 72 | 13(18.06 %) | | | |
| Agro-ecology | Highland | 23 | 2 (8.69 %) | | | |
| | Midland | 27 | 9(33.30%) | 2 | 6.816 | 0.033 |
| | Lowland | 22 | 2(9.09%) | | | |
| Over all Prevalen | ce | 72 | 13(18.06%) | | | |

Source: Survey result, 2022

Infestation of varroa mite

The current result indicated that varroa mite infestation in workers honeybees' were higher in transitional beehives (91.67%) followed by modern beehives (77.5%) and the least infestation was found in traditional beehive (47.37%) with overall mean (71.83%). The result indicates that infestation of varroa mites is less likely to impact honeybee health and its production in the study areas. Similarly, Begna *et al.* (2016) state that the level of honeybee population and their activities in nectar collections did not affected by the varroa mites infestation. The result also indicated that varroa mites' infestation was more common in lowland (81.81%) followed by mid highland (76.92%) while the lowest was found in highland agro ecology (56.52%) (Table13).

The overall varroa mite infestation in honeybee broods was observed 21.78%. However, the distribution was ranges from 9.09% to 50% in brood. The present result indicated that varroa mite infestation in honeybee broods was higher in traditional behives (35.71%) followed by transitional behives (25.00%) and modern behive (14.29%). The result also indicated that varroa mites' infestation in honeybee broods' were more common on midland (50%) followed by lowland (10.0%) while the lowest was found in highland agro ecology (9.09%) %) (Table13).

| Variable | Category | Adult sample coloni | es infestation (%) | brood samples | infestation (%) |
|-----------------------------|---------------|---------------------|--------------------|---------------|-----------------|
| Bee hive types | Movable frame | 42 | 31(77.5%) | 28 | 4(14.29 %) |
| | Transitional | 12 | 11(91.67%) | 4 | 1(25 %) |
| | Traditional | 19 | 9(47.37%) | 14 | 5(35.71 %) |
| Over all infestation | | 71 | 51(71.83 %) | 46 | 10 (21.74%) |
| Agro-ecology | Highland | 23 | 13 (56.52 %) | 22 | 2(9.09%) |
| | Midland | 26 | 20(76.92%) | 14 | 7(50 %) |
| | Lowland | 22 1 | 8(81.81%) | 10 | 1(10 %) |
| Over all infestation | | 72 | 51(71.83 %) | 46 | 10 (21.74% |

Table 13. Infestation of adult and varroa mite brood

Source: Survey result, 2022

Prevalence of Bee lice and Associated Risk Factors

The presented result indicated that the prevalence of bee lice was higher in transitional beehives (16.67%) followed by traditional beehives (33.33%) and modern beehives (26.19%) (Table14). The study result showed that the prevalence of *bee lice* was observed 31.81 % in lowland followed by 25.96 % in midland while the lowest was found 21.74 % in highland. The current study agrees with report Nega *et al.* (2019). Table 14. Prevalence of bee lice and associated risk factor

| Variables | Category | No. of sampled colonies | Prevalence (%) | df | x ² | p-values |
|---------------------|-------------------------------|-------------------------|-------------------------|----|----------------|----------|
| Beehives types | Movable frame Transitional | 42 12 | 11(26.19%) 2(16.67%) | 2 | 1.032 | 0.597 |
| Beenives types | Traditional | 12 | 6(33.33%) | 2 | 1.052 | 0.577 |
| Agro-ecology | Highland | 23 | 5(21.74 %) | 2 | 0.502 | 0.742 |
| | Midland Lowland | 27 22 | 7(25.96%) 7(31.81%) | 2 | 0.593 | 0.743 |
| Over all prevalence | e | 72 | 19(26.39 %) | | | |

Source: Survey result, 2022

Prevalence of Wax moth and Associated Risk Factors

The current result showed that prevalence of Wax moths was highest in modern behives (45.24%), followed transitional behives (41.67%) and the lowest prevalence was observed in traditional behives (38.89%). This might be because of poor management practices by beekeepers in modern behives and transitional behives like no replace of old combs and no remove of queen excluder throughout the year as we had observed during sample collections. Moreover, concerning agro-ecologies high prevalence of *wax moth* was observed %) in the highland (48.83 %) followed midland (40.74%) by while the lowest was observed in lowland 31.81%) (Table15). Table 15: Prevalence of Wax moth and Associated Risk Factors

| Variables | Category | No. of sampled colonies | Prevalence (%) | df | x ² | p-values |
|-------------------|---------------|-------------------------|----------------|----|----------------|----------|
| | Movable frame | 42 | 19(45.24%) | | | |
| Beehives types | Transitional | 12 | 5(41.67%) | 2 | 0.219 | 0.897 |
| | Traditional | 18 | 7(38.89%) | | | |
| Agro-ecology | Highland | 23 | 11(48.83 %) | | | |
| | Midland | 27 | 11(40.74%) | 2 | 0.314 | 0.855 |
| | Lowland | 22 | 7(31.81%) | | | |
| Over all prevaler | nce | 72 | 29(40.27 %) | | | |

Source: Survey result, 2022

Prevalence of Small Hive Beetles and Associated Risk Factors

Small hive beetle (SHB) is a nest parasite of honey bees (*Aphis mellifera* L.) colonies native to Sub-Saharan Africa and is considered a minor pest of honeybees. The overall prevalence of small hive beetles was observed 54.22% across agro-ecologies. The highest prevalence of small hive beetles was detected in the lowland (63.6%) followed by the midland (51.90%) while the lowest was observed in the highland (48.80%) in the study districts (Table 16). This might be due to this pest favors to hot temperature and low relative humidity along maize and sorghum cultivated land and livestock rearing area. This finding agreement with Nega *et al.* (2019) who stated that variation in agro-ecologies may be attributed to different factors such as ecological variability, season, and management aspects.

Regarding hive types, the result indicated that high prevalence of SHB was observed in modern beehives (61.90%) followed by transitional beehives (50.00%) while the lowest prevalence was observed in traditional

beehives (38.90%). This is probably due to poor management practices of beekeepers had on modern and transitional hives in the study districts during the study period.

| Category | No. of sampled colonies | Prevalence (%) | df | x ² | p-values |
|---------------|---|---|--|--|--|
| Movable frame | 42 | 26(61.90%) | | | |
| Transitional | 12 | 6(50.00%) | 2 | 2.789 | 0.248 |
| Traditional | 18 | 7(38.90%) | | | |
| Highland | 23 | 11(47.80 %) | | | |
| Midland | 27 | 14(51.90%) | 2 | 1.225 | 0.542 |
| Lowland | 22 | 14(63.6%) | | | |
| | Movable frame Transitional Traditional Highland Midland | Movable frame42Transitional12Traditional18Highland23Midland27 | Movable frame 42 26(61.90%) Transitional 12 6(50.00%) Traditional 18 7(38.90%) Highland 23 11(47.80 %) Midland 27 14(51.90%) | Movable frame 42 26(61.90%) Transitional 12 6(50.00%) 2 Traditional 18 7(38.90%) 11(47.80%) Highland 23 11(47.80%) 2 Midland 27 14(51.90%) 2 | Movable frame 42 26(61.90%) Transitional 12 6(50.00%) 2 2.789 Traditional 18 7(38.90%) 2 2.789 Highland 23 11(47.80 %) 2 1.225 |

Table 16: Prevalence of Small hive beetles and Associated Risk Factors

Source: Survey result, 2022

Conclusions and Recommendations

The present study identified major challenges of beekeeping practices in the study areas were pests and predators, shortage of bee forage, absconding, application of agrochemicals, diseases, mass death of bee colonies during the study period. Majority of the beekeepers practicing traditional beekeeping which are more face for these problems. Furthermore, there is lack of knowledge and skill gap about modern beekeeping equipment how to use and practices in the study areas. The study finding revealed that the major honeybee pest and predators according to important order were ranked. These pests and predators including, honey badger, spider, ant, wax moth, bee-eater birds and small hive beetles and monkeys were declared by respondents. Honey badger is the most dangerous predator for honeybee diseases and pests in the study areas were Nosema apis, Amoeba, varroa mites, bee lice, wax moth, and small hives beetle. However, current study were not detected AFB, EFB, and trachea mites during laboratory analysis. In conclusion, across agro-ecologies and beehive types were identified as risk factors for prevalence of honey bee diseases and pests in the study areas.

Based on the above conclusion the following recommendations are forwarded;

- Awareness creation should be needed in terms of technical knowledge and skills that could capacitate the knowledge of beekeepers to benefit them from the apiculture sub-sector,
- Beekeepers should be maintain strong and healthy honeybee colonies via proper seasonal colony management practices from diseases, pests and predators,
- Beekeepers should avoid contamination of bee equipment, avoid transfer of infected combs to healthy colony, avoid common feeding of honeybee colonies which spread diseases and pests transmission,
- There is a great need to develop scientifically mechanisms for controlling of pests and predators,
- Further, study on seasonal prevalence and outbreak of honeybee diseases and pests could be very important.

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