

(Original Research)

Molecular Evidence of *Ancylostoma* spp. Infection among Animals in Selected Agricultural Farms in Laguna and Quezon Province, the Philippines: Implications to Food Security and Safety

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Abstract

Background: In achieving food security, the role of animals as source of infection is particularly neglected, especially in agricultural farms where focus is on the organic farming system. This study aimed to assess the role of animals in the perpetuation of hookworm infection in selected agricultural farms in Laguna and Quezon Province, the Philippines. **Methods:** A total of 161 fecal samples were collected in agricultural farms in Laguna and Quezon. DNA was extracted and subjected to real-time PCR to detect *N. americanus* and *Ancylostoma* spp. **Results:** High prevalence of *Ancylostoma* spp. (20.5%) among animals with highest prevalence seen in companion animals (30.0%) while no *N. americanus* were detected. Significant variability among farms is observed implying differences in agricultural practices. Deworming and containment status of animals as well as awareness of owners/managers decreases the likelihood of hookworm infection. **Conclusion:** Animals serve as sources of parasite contamination for fresh produce as well as reservoirs for human infection threatening food safety and security. Reviewing existing guidelines

for good agricultural practices to include the role of animals as well as parasites in contamination is needed to accelerate control and elimination of this parasitic infection. An interdisciplinary approach may be needed to move forward in achieving food security.

Keywords

Food security, Food safety, Hookworm, *Ancylostoma* spp.

1. Introduction

Food insecurity or the lack of regular access to nutritious and healthy food continues to threaten the health of many people around the globe especially in poor and developing countries, such as the Philippines [1]. It is estimated that six out of ten Filipinos are food insecure [2]. Nutritious and healthy food are necessary for normal growth and development, the lack thereof contributes to several morbidities such as undernutrition, malnutrition, stunting, and wasting creating a cascade of several economic, societal, and health

problems [3]. The United Nations recognizes the severity of this problem as such the Sustainable Development Goals (SDGs) aims to achieve zero hunger and food security by 2030 [4]. Strengthening agriculture through increasing crop yield and decreasing crop loss is among the strategies used to achieve food security. This has been made possible by adopting a conventional farming system where chemical fertilizers and pesticides are being used [5,6]. However, these synthetic fertilizers and pesticides pollute the environment making it an unsustainable strategy to achieve SDG [5].

To achieve the same effects without the harmful chemicals, organic farming was introduced. In this traditional type of farming, nutrients existing in the farm are utilized to enhance and support growth and development of crops. Existing nutrients include animal manure being used as fertilizers [5,7]. However, this organic manure can cause diseases for it carries several pathogens [8]. Strategies to mitigate food insecurity, such as utilizing untreated organic manure, inadvertently threaten food safety by facilitating the transmission of zoonotic pathogens, including parasites [9,10].

Among these parasites, hookworm became notorious in both veterinary and public health as it infects an estimated 438.9 million people globally [11]. Hookworms may be transmitted either by skin penetration or ingestion. In the Philippines, hookworm along with other soil-transmitted helminths remains a public health burden due to its high infectivity rate as well as heavy intensity infection is associated with stunting, malnutrition, and anemia among others [12,13].

Animals play a role in the perpetuation of these parasites. They are potential reservoir host of parasitic zoonosis and considered as an overlooked factor that can contribute to the burden and transmission of hookworms due to their close interaction to humans and their use in the agriculture sector as a livelihood, resulting in a broader spectrum of transmission, parasitic persistence, loss of productivity and income, and food and work safety of both farmers and consumers [14,15]. This neglected contribution of animals amplifies the risk of environmental contamination in an integrated farming system where both crops and animals are produced in one

farm. Thus, this study aimed to assess the role of animals in the perpetuation of hookworm infection in selected agricultural farms in Southern Luzon, Philippines.

2. Materials and Methods

2.1. Study Sites

This cross-sectional study was conducted in selected farms in Laguna and Quezon, Philippines. Both provinces are known to have medium to large-scale integrated farm systems. A total of 11 farms were included as study sites with six farms from Laguna and five farms from Quezon. Farms were selected based on the following criteria: (1) consent of farm owners; (2) practicing integrated farming of crops and livestock; and (3) small-to-medium holder farms with a land area of at least 500 square meters. Selection of farms as study sites was properly coordinated with the Provincial and Municipal Agricultural Offices of Laguna and Quezon.

2.2. Sample and Data Collection

Approximately 50g of naturally passed fecal samples were collected from animals in selected agricultural farms. Sample freshness was determined by either observing the animal's defecation or noting the presence of a moist sheen on the sample surface. Collected samples were immediately transported to the field laboratory via cold chain for processing and examination. Animal conditions and care practices such as animal sex, age, containment status, deworming status, water source, and contact with other animals were also documented.

Furthermore, a survey questionnaire regarding the farming practice and sanitary conditions of the farms as well as knowledge on parasitic infection was also administered to the owner or the manager of each farm. Prior to the actual data collection, the survey questionnaire was pretested in an integrated farm in Batangas province and was revised accordingly.

2.3. DNA Extraction

Genomic DNA from collected fecal samples was extracted using Nucleospin® Soil DNA isolation kit (Macherey – Nagel GmbH & Co. KG) following the manufacturer's instructions.

DNA purity and concentration were confirmed using Multiskan SkyHigh UV-Vis microplate spectrophotometer (Thermo Scientific TM). Extracted DNA samples were stored at -20°C until further downstream analysis.

2.4. Real-time PCR

Multiplex real-time PCR assays were used to detect the presence of *Necator americanus* and *Ancylostoma* spp. on the extracted DNA samples. For *N. americanus* detection, the following primers and hydrolysis probes were used: Fwd_N. amer: 5'-CTGTTTGTCTGAACGGTACTTGC-3' and, Rev_N. amer: 5'-ATAACAGCGTGCACATGTTG-3' and 5'-[6FAM]CTGTACTACGCATTGTATAC[MGBNFQ]-3', which target the 101bp-ITS2 region specific for *N. americanus*. On the other hand, the 72bp-ITS2 region is specific for *Ancylostoma* spp. was amplified for its detection using the following primers and probes: Fwd_Ancy: 5'-GAATGACAGCAAACCTCGTTGTTG-3' and, Rev_Ancy: 5'-ATACTAGCCACTGCCGAAACGT-3' and 5'-[VIC]ATCGTTTACCGACTTTAG [MGBNFQ]-3' [16] (Verweij *et al.*, 2007).

Assays were run in 10 µL reaction volume which contains 5 µL mastermix (QuantiTect Multiplex PCR NoROX Kit, Qiagen), 10 µM of each primer and probes and, 2 µL DNA template, and 1.9µL nuclease-free water. Magnetic Induction Cyclor (MIC) qPCR cyclor (Bio Molecular Systems, Australia) was used for the assay with an optimized amplification profile of 15 min at 95°C followed by 40 cycles of 60 secs at 94°C, 60 secs at 60°C. Positive controls containing the targeted gene fragments for *N. americanus* and *Ancylostoma* spp. as well as negative control using nuclease-free water were also included in each run for quality control purposes. A Cq value of ≤35 was used as a cut-off to distinguish between positive and negative samples based on previously published literature utilizing the same assay [11].

2.5. Data Processing and Analysis

Data gathered were encoded using Microsoft Excel 2010. As part of quality control, all encoded data were re-checked for discrepancies and resolved as necessary. Data analysis was carried using STATA version 17 (STATA 17®, Stata Corp, Texas, USA). Prevalence of hookworm per animal group and per farm was determined. Descriptive statistics was used for the farming practices and

sanitary conditions of agricultural farms. Furthermore, point biserial correlation was used to identify the relationship of farming practices and sanitary conditions as well as knowledge on parasitic infection in each farm.

Multiple logistic regression was used to identify predictive factors associated with hookworm infection. Bivariate analysis of outcome and variables was carried out prior the regression analysis. Significant variables were used in the regression model. Backward stepwise multiple logistic regression analysis was done to identify predictive variables in the final model. In all analyses, a p-value of less than 0.05.

2.6. Ethical Considerations

The study protocol was reviewed and approved by the University of the Philippines (UP) Manila Research Ethics Board (UPMREB 2018-201-01) and UP Los Baños Institutional Animal Care and Use Committee (UPLB 2021-039). Consultation with the local government units of Laguna and Quezon was conducted prior to the implementation of the study. Informed consent from farm owners was obtained before the conduct of the interviews. Confidentiality was ensured through a unique numerical identifier assigned in each farm as well as animals and any personal information that could identify the farms were removed following the Data Privacy Act of 2012.

3. Results

A total of 11 agricultural farms were included in this study. A short description of each farm was included in Table 1. In Laguna, most of the farms were practicing organic farming with only one practicing conventional farming system. On the other hand, selected farms in Quezon practice both organic and conventional farming systems. Of all those practicing the organic farming system, only one farm was certified by the Organic Certification Center of the Philippines (OCCP). All the selected farms, regardless of whether organic or conventional, use animal manure as fertilizers. Presence of animals, both stray and livestock, is also noted in all the farms.

Table 1. Description of selected agricultural farms in Laguna and Quezon Province, Philippines.

Farm Type		Brief Description of Farms
Laguna		
1	Practicing Organic	Integrated farming; Presence of stray dogs, cats, chickens; Use of vermicomposted pig manure; Use of surface water source; Use of the same farming tools for animals and cropping area
2	Practicing Organic	Integrated farming; Use of untreated pig manure; Use of surface water source; Use of the same farming tools for animals and cropping area
3	Practicing Organic	Integrated farming; Eco-tourism farm (lettuce- and strawberry-picking activity for tourists); Use of dried chicken manure as fertilizer; Chicken and dog are leashed but in contact with stray dogs and cats; Use of the same farming tools for animals and cropping area
4	Conventional	Companion animals exposed to stray animals; Nearby spring as water source
5	Practicing Organic	Integrated farming; Agriculture learning site and eco-tourism farm with recreational pools; Presence of stray dogs and cats; Goat is leashed but exposed to other animals; Use of dried pig, cattle, goat, and chicken manure as fertilizer; Use of groundwater
6	Practicing Organic	Eco-tourism farm with lodging rooms and toilet near cropping area; Presence of stray dogs, chickens, and cats; Use of groundwater
Quezon Province		
7	Practicing Organic and Conventional	Integrated farming with both organic and conventional farming areas; Agriculture learning site; Presence of stray dogs, cats, chicken; Use of vermicomposted cattle and chicken manure; Use of surface water source
8	Practicing Organic and Conventional	Integrated farming with both organic and conventional farming areas; Use of dried cattle, goat, chicken manure; Use of surface water source; Free-range sheep; Possible farm run-offs with the fecal matter due to poor drainage systems in some animal enclosures
9	Practicing Organic	Integrated farming, agriculture learning site, and eco-tourism farm; Ruminants are leashed but located near cropping areas; Free-range chicken, cats, and dogs; Use of vermicomposted chicken and rabbit manure; Use of groundwater source, use of the same farming tools for animals and cropping area
10	Conventional	Livestock farm with limited crops (fruit-bearing trees and okra), presence of stray cats and dogs; Use of dried goat and rabbit feces; Use of groundwater source
11	Certified Organic	Integrated farming, agriculture learning site and eco-tourism farm

A total of 161 fecal samples from animals were collected from the 11 agricultural farms, 54 of which were from Laguna while the remaining 107 were from Quezon. Upon molecular analysis, only *Ancylostoma* spp. was detected with an overall

prevalence of 20.5%, whereas no samples tested positive for *N. americanus*. Highest prevalence was observed among companion animals with 30.0%, followed by swine (25.0%), poultry (18.6%), and ruminants (10.5%) (Table 2).

Table 2. Prevalence of *Ancylostoma* spp. among animals in selected agricultural farms in Laguna and Quezon Province, Philippines.

Animal Type	Prevalence of <i>Ancylostoma</i> spp.					
	Laguna		Quezon Province		Overall	
	N	Positive (%)	N	Positive (%)	N	Positive (%)
Poultry	23	3 (13.0)	36	8 (22.2)	59	11 (18.6)
Ruminants	5	0	33	4 (12.1)	38	4 (10.5)
Swine	13	1 (7.7)	11	5 (45.4)	24	6 (25.0)
Companion Animals	13	6 (46.2)	27	6 (22.2)	40	12 (30.0)
Overall	54	10 (18.5)	107	23 (21.5)	161	33 (20.5)

Table 3 summarizes the results of the bivariate analysis of animal condition and care practices and *Ancylostoma* spp. infection.

Prevalence of *Ancylostoma* spp. is significantly different among animal groups, with the highest prevalence observed in companion animals at

Table 3. Bivariate analysis of animal conditions and care practices and *Ancylostoma* spp. infection among animals in selected agricultural farms in Laguna and Quezon, Philippines.

Variables	Frequency	n (%)	X ²	p value
Overall (N=161)				
<u>Province</u>				
Laguna	54	10 (18.5)	0.195	0.659
Quezon	107	23 (21.5)		
<u>Animal group</u>				
Poultry	59	11 (18.6)	34.710	0.001*
Ruminants	38	4 (10.5)		
Swine	24	6 (25.0)		
Companion animals	40	12(30.0)		
<u>Deworming of animals</u>				
No	108	27 (25.0)	4.082	0.043*
Yes	53	6 (11.3)		
<u>Containment</u>				
Penned/Leashed	130	21 (16.1)	7.815	0.005*
Free roaming	31	12 (38.7)		
<u>Type of food</u>				
Organic	110	28 (25.4)	5.238	0.073
Commercial	41	4 (9.8)		
Mix	10	1 (10.0)		
<u>Source of water</u>				
Surface	77	12 (15.6)	2.186	0.139
Ground	84	21 (25.0)		
<u>Contact with other animals</u>				
Yes	85	25 (29.4)	8.782	0.003*
No	76	8 (10.5)		
Poultry Animals (N=59)				
<u>Province</u>				
Laguna	23	3 (13.0)	0.779	0.377
Quezon	36	8 (22.2)		
<u>Deworming of animals</u>				
No	35	6 (17.1)	0.128	0.721
Yes	24	5 (20.8)		
<u>Containment</u>				
Penned/Leashed	43	4 (9.3)	9.123	0.003*
Free roaming	16	7 (43.7)		
<u>Type of food</u>				
Organic	29	7 (24.1)	1.135	0.287
Commercial	30	4 (13.3)		
Mix	0			
<u>Source of water</u>				
Surface	26	3 (11.5)	1.547	0.214
Ground	33	8 (24.2)		
<u>Contact with other animals</u>				
Yes	36	10 (27.8)	5.079	0.024*
No	23	1 (4.3)		
<u>Sex</u>				
Male	40	6 (15.0)	1.087	0.297
Female	19	5 (26.3)		
<u>Age Group</u>				
0-11 days	13	1 (7.7)	1.344	0.511
12-21 days	10	2 (20.0)		
More than 21 days	36	8 (22.2)		

Ruminants (N=38)				
<u>Province</u>				
Laguna	5	0	0.677	0.410
Quezon	33	4 (12.1)		
<u>Deworming of animals</u>				
<u>Deworming of animals</u>				
No	27	4 (14.8)	1.821	0.177
Yes	11	0		
<u>Containment</u>				
Penned/Leashed	28	3 (10.7)	0.004	0.950
Free roaming	10	1 (10.0)		
<u>Type of food</u>				
Organic	38	4 (10.5)	-	-
Commercial	-	-		
Mix	-	-		
<u>Source of water</u>				
Surface	20	2 (10.0)	0.124	0.911
Ground	18	2 (11.1)		
<u>Contact with other animals</u>				
Yes	32	4 (12.5)	0.838	0.360
No	6	0		
<u>Sex</u>				
Male	15	3 (20.0)	2.362	0.124
Female	23	1 (4.3)		
<u>Age Group</u>				
Less than 2 years	13	0	2.336	0.311
2 to 3 years	12	2 (16.7)		
More than 3 years	13	2 (15.4)		
Swine (N=24)				
<u>Province</u>				
Laguna	13	1 (7.7)	4.532	0.033*
Quezon	11	5 (45.4)		
<u>Deworming of animals</u>				
No	20	6 (30.0)	1.600	0.206
Yes	4	0		
<u>Containment</u>				
Penned/Leashed	24	6 (25.0)	-	-
Free roaming	-	-		
<u>Type of food</u>				
Organic	13	6 (46.1)	6.769	0.009*
Commercial	11	0		
Mix	-	-		
<u>Source of water</u>				
Surface	13	1 (7.7)	4.532	0.033*
Ground	11	5 (45.4)		
<u>Contact with other animals</u>				
Yes	-	-	-	-
No	24	6 (25.0)		
<u>Sex</u>				
Male	11	4 (36.4)	1.399	0.237
Female	13	2 (15.4)		
<u>Age Group</u>				
Less than 3 months	-	-	0.348	0.555
3 to 6 months	1	0		
More than 6 months	23	6 (26.1)		
Companion Animals (N=40)				
<u>Province</u>				
Laguna	13	6 (46.1)	2.393	0.122
Quezon	27	6 (22.2)		

<u>Deworming of animals</u>				
No	26	11 (42.3)	5.358	0.021*
Yes	14	1 (7.1)		
<u>Containment</u>				
Penned/Leashed	35	8 (22.9)	6.803	0.009*
Free roaming	5	4 (80.0)		
<u>Type of food</u>				
Organic	30	11 (36.7)	2.540	0.111
Commercial	-	-		
Mix	10	1 (10.0)		
<u>Source of water</u>				
Surface	18	6 (33.3)	0.173	0.677
Ground	22	6 (27.3)		
<u>Contact with other animals</u>				
Yes	17	11 (64.7)	16.958	0.000*
No	23	1 (4.3)		
<u>Sex</u>				
Male	23	8 (34.8)	0.590	0.443
Female	17	4 (23.5)		
<u>Age Group</u>				
Less than a year	12	2 (16.7)	1.451	0.228
1 to 5 years	28	10 (35.7)		
More than 5 years	-	-		

* significant at $p < 0.05$

30.0% ($p = 0.001$). Significantly lower prevalence is observed in dewormed animals compared to those not dewormed ($p=0.043$), however, this is not observed among poultry animals. Penned/leashed animals showed significantly lower prevalence compared to free-roaming animals which was observed across animal groups ($p < 0.05$). Another significant variable is the contact with other animals, where animals sharing a space with other animals have significantly higher prevalence compared to those that do not. Furthermore, among swine, the type of food and source of water contributes to *Ancylostoma* spp. infection where swine that feeds on organic food and drinks on ground water have significantly higher prevalence ($p=0.009$ and $p=0.033$, respectively). This was not observed in other animal groups.

Table 4 shows the best logistic regression model for predicting factors contributing to *Ancylostoma* spp. infection. Swine and companion animals have a higher likelihood of being infected with *Ancylostoma* spp. compared to poultry

animals. On the other hand, restricted contact with other animals reduces the likelihood of being infected with *Ancylostoma* spp. in selected agricultural farms.

Point biserial correlation of the KAP only revealed three factors with significant relationship to *Ancylostoma* spp. infection. The awareness of farmers that manure is a source of parasite contamination to crops is negatively correlated to *Ancylostoma* spp. prevalence in the farm. Similarly, awareness of human-to-animal transmission and vice versa are also negatively correlated to *Ancylostoma* spp. prevalence in the selected agricultural farms (Table 5).

As efforts are directed toward achieving food security through sustainable organic farming practices, food safety concerns may arise, particularly in agricultural farms where integrated crop-livestock farming systems are observed. The potential role of animals in the contamination of produce and perpetuation of hookworm transmission in humans requires

Table 4. Predictive factors contributing to *Ancylostoma* spp. infection among animals in selected agricultural farms in Laguna and Quezon, Philippines.

Outcomes/Variables	Odds Ratio	95% CI	p value
Animal group (baseline poultry animals)			
Ruminants	0.3	0.1-1.2	0.093
Swine	17.8	2.6-120.8	0.003*
Companion animals	3.8	1.2-12.0	0.021*
No contact with other animals	0.4	0.0-0.2	0.000*

* significant at $p < 0.05$

further investigation. The extent of environmental contamination and direct transmission to humans remains unclear because only animal fecal samples were analyzed.

farmers and farmworkers who, daily, are in contact with the soil and or manure fertilizers [19]. As observed in some of the farms, wearing protective gears such as gloves and boots is not a

Table 5. Association of factors to *Ancylostoma* spp. prevalence among animals in selected farms in Laguna and Quezon, Philippines.

Factors	Pearson r	p value
Manure as fertilizers		
Farms use manure as fertilizers	0.340	0.307
Farms treat manure before application	0.222	0.511
Water source		
Surface water (spring, creek, river, pond)	-0.225	0.505
Ground water (pump, artesian well)	0.225	0.505
Sanitation and hygiene practices		
Use of protective gears (gloves, boots, etc.)	-0.169	0.620
Cleaning of containers, crates, farming tools	-0.096	0.780
Washing of fresh produce	-0.459	0.155
Washing of farm tools	-0.550	0.080
Handwashing before work	-0.089	0.794
Handwashing after work	<i>a</i>	<i>a</i>
Availability of toilet facility	<i>a</i>	<i>a</i>
Agriculturist/Veterinarian inspection	0.559	0.074
Presence of stray animals	<i>a</i>	<i>a</i>
Knowledge on parasite transmission		
Transmission of parasites from manure to crops	-0.696	0.017*
Transmission of parasites from animals to humans	-0.839	0.001*
Transmission of parasites from humans to animals	-0.839	0.001*

* significant at $p < 0.05$

a cannot be computed due to constant variable

Overall, the results showed relatively high prevalence of *Ancylostoma* spp. among animals in the selected agricultural farms. The presence of hookworm infection among animals may not only affect the host itself but may also contribute to the potential contamination of fresh produce in the agricultural farms as well as the infection among farmers and other farmworkers [10,17,18]. The free-roaming animals in some of the selected farms in addition to their natural propensity to open defecate are at high risk of contaminating not only the soil but also the vegetable produce. The term 'organic farming' usually connotes that produce is ready to be served directly from farm to table and that they are free from any harmful chemical contaminants. However, the possible parasite contamination of these fresh produce highlights the need for strengthening health education campaigns focusing on the proper preparation of foods even when they are said to be organic.

Furthermore, this also puts farmers and other farmworkers in agricultural farms at higher risk for contracting hookworm infection. In reference to the transmission of hookworms which is both skin penetration and ingestion of infective larvae, the usual handwashing may not be as effective in preventing hookworm infection especially for

common practice, thus may increase the likelihood of larval skin penetration. Including the risk of parasitic infection in formulating a standard guideline for good agricultural practices may be needed to address this issue among at risk populations.

Logistic regression analysis also revealed that companion animals such as cats and dogs are more at risk in being infected with *Ancylostoma* spp. due to their behavior compared to poultry. These may be because of the behavior of animals and host specificity of parasites. Movement of poultry animals are usually restricted as observed in the selected farms compared to companion animals. This limits the latter's exposure to parasites. Furthermore, although *Ancylostoma* spp. has a wide host range, they are known to infect mammals. Hence, the higher prevalence among companion animals and swine compared to poultry is common. The significantly highest prevalence observed in companion animals is alarming as they pose the greatest transmission risk to humans as companion animals have the closest contact with humans [20].

Results also revealed significantly lower prevalence of *Ancylostoma* spp. in dewormed

animals compared to those that were not, showing the effectiveness and importance of deworming in controlling parasitic infections. However, in the strictest sense of organic farming, synthetic anthelmintics are prohibited [5,6]. This was not strictly implemented in the farms included in this study as some reported the use of synthetic ones. On the other hand, organic alternatives are sometimes used which may not be as effective as synthetic anthelmintics whose efficacy has been clinically proven to eradicate parasitic infection among animals. Alternative parasite management strategies that are both effective and consistent with organic farming principles should be evaluated. While the goal of achieving organic practice is the same, differences in farm management and sanitary practices among the selected farms were observed, which may influence the occurrence of *Ancylostoma* spp. infection and further highlight the need for standardization of agricultural practices.

Furthermore, free-roaming animals demonstrated significantly higher prevalence compared to penned/leashed animals. Although leashed, some animals maintained contact with other animals, potentially serving as a source of infection. This interaction likely contributed to the higher parasite prevalence compared to animals kept in enclosed spaces without external contact. This emphasizes the need for a proper enclosure of animals to minimize infection risk and improve control of parasitic infection among animals. This is also supported by the results of logistic regression analysis where enclosure without contact to other animals decreases risk of being infected [21].

Results of point biserial correlation revealed a negative correlation between animal prevalence and the awareness of farm owners. As awareness increases, the prevalence of infection decreases. This emphasizes the importance of health education focusing on increasing awareness and promoting behavioral change. Further contextualizing health education campaigns for the control of parasitic infection to fit the agricultural farms set up may be more beneficial for farmers and other farmworkers [22].

5. Conclusions

The findings of the study showed the role of animals in *Ancylostoma* spp. infection in selected

agricultural farms. *Ancylostoma* spp. remains one of the persisting soil-transmitted helminth infections globally, which may be attributed to its zoonotic potential and the role of animals as infection reservoirs that initiate transmission in agricultural settings. The high prevalence of *Ancylostoma* spp. in farm animals in the present study highlights their role as potential reservoirs for human infection and underscores the significant risk of parasite contamination for fresh produce cultivated in integrated crop-livestock farming systems. The lack of standard agricultural practices led to differences in the implementation of farming systems which contributed to the transmission and, ultimately, perpetuation of parasitic infection. Revisiting existing guidelines for good agricultural practices to include the role of animals as well as parasites in contamination is needed to accelerate control and elimination of these parasitic infections. Results of the study also showed the necessity for health education campaigns to increase awareness among farmers and decrease the risk of infection. The study highlighted that achieving food security and safety may not be as straightforward as moving from one farming system to another but rather an interdisciplinary approach accounting for the contributions of the environment, animals, and humans.

Availability of Data and Materials

The data that support the findings of this study are available upon reasonable request.

Author Contributions

Conceptualization, A.I.A., F.N.M., S.R.E., J.R.M., and V.V.P.; Methodology, A.I.A., F.N.M., S.R.E., J.R.M., and V.V.P.; Investigation, A.I.A., F.N.M., and S.R.E.; Writing – Original Draft, A.I.A. – Review & Editing, A.I.A., F.N.M., S.R.E., J.R.M., and V.V.P.; Funding Acquisition, J.R.M., and V.V.P.; Resources, A.I.A., J.R.M., and V.V.P.; Supervision, V.V.P.

Ethics Approval and Consent to Participate

Ethical approval from the University of the Philippines Los Baños Institutional Animal Care and Use Committee (UPLB IACUC) and the University of the Philippines Manila Research Ethics Board (UPM REB) was obtained prior to the conduct of the study. Both protocols for human participant involvement and animal subjects were

found to conform to the expectations of the University regarding ethical standards and good practices in human and animal research.

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Conflict of Interest

The authors declare no conflict of interest.

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