



## Epidemiology of free-living amoebae in the Philippines: a review and update

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### ABSTRACT

Free-living amoebae (FLA) are considered environmental pathogens and thus pose a public health threat. Their ubiquity in natural sources may magnify the potential severity of health outcomes in the future. However, less attention was given despite several probable public health risks that arise from the presence of pathogenic strains in the environment. Here, we provide epidemiological data based on investigations involving the distribution and occurrence of free-living amoebae in the Republic of the Philippines. This aims to connect data of fragmented studies of these organisms and provide potential roadmaps in FLA research in the country. The majority of the reviewed articles ( $n = 19$ ) focused on characterization studies (36.8%; 7/19) while environmental isolation and isolation from biological samples had an equal frequency of 31.6% (6/19) each. There is a great disparity between the established ubiquity in environmental sources and the number of cases of FLA infections in the country. FLA-related research in the Philippines is still in its inceptive stage with several gaps to fill, which can be used to formulate policy briefs in the future regarding its isolation, identification, diagnosis, therapeutic management, and control of FLA infections in the country.

### KEYWORDS

*Acanthamoeba*;  
epidemiology; free-living  
amoeba; *Naegleria*;  
Philippines

### Introduction

Studies on free-living amoebae (FLA) have in recent decades gained traction in the field of protozoology and parasitology. Known for their ubiquity, FLAs have been isolated from various environmental matrices, such as soil, fresh and brackish water, wastewater, hot springs, dust, and others [1–6]. Aside from environmental sources, FLAs were also reported from cooling towers, air conditioning systems, bromeliad plants, dental water supplies, and are known to contaminate contact lens and lens fluids [7]. In addition, FLA can harbor pathogenic microorganisms within its cytoplasm, enabling them to persist and eventually be transmitted to a broad spectrum of biological hosts via multiple transmission routes. Due to this, FLA is dubbed as ‘Trojan horses’ of the microbial community [8,9]. Prokaryotes persisting within FLA have been called amoebae-resistant bacteria (ARB) by some researchers [10], while most recently, the collective term ‘endocytobionts’ has been used to describe the variety of microorganisms that multiply within an FLA host [11]. This term may be more appropriate considering that FLA, in general, can also internalize viruses [12], fungi [13], and protozoa [4,14,15].

Among the FLAs, the genera belonging to *Naegleria*, *Acanthamoeba*, *Balamuthia*, and *Sappinia* are considered by the World Health Organization

(WHO) as medically important due to the morbidity or mortality reports in humans [16]. The route of cerebral infections for pathogenic FLA, in particular *Naegleria* spp., is initiated by the entry of the amoeba via the nasal cavity usually upon inhalation of contaminated water [17]. Upon reaching the brain via the cribriform plate, FLAs can mediate cytopathic effects resulting in the inflammation of the brain known as meningitis [18]. Depending on the FLA species or genotype and type of infection, conditions have been referred to as Primary Amoebic Meningoencephalitis (PAM) for *Naegleria* spp. infections [19], Granulomatous Amoebic Meningoencephalitis (GAE) for *Acanthamoeba* spp. infection [20], *Balamuthia* Amoebic Encephalitis (BAE) for *Balamuthia mandrillaris* infection, and *Sappinia* Amoebic Encephalitis (SAE) for *Sappinia* spp. infections [21]. Clinical conditions have almost equal morbidity to mortality ratio due to the rapid progression of the disease following the onset of symptoms [22]. Further, the symptoms presented by FLA-related meningitis mimic viral and bacterial forms, thus, making diagnosis and management of the disease challenging for clinicians and almost always leads to death [23]. Among the FLAs, pathogenic genotypes of *Acanthamoeba* spp. can inflict extra-cerebral infections like *Acanthamoeba* keratitis, and in rare cases, disseminated cutaneous infection [24–27].

The tenacity of FLA to proliferate in harsh ecosystems, and survive desiccation or disinfection make these organisms an important emerging waterborne, foodborne, and airborne protozoan pathogen of our time [11]. Considering that the overall global burden of FLA-related human infections is relatively low and infections from FLA are still rare [28,29], the almost equal proportion of morbidity to mortality ratio of reported cases, especially those causing a meningitis-like condition and the lack of effective therapeutic regimen, necessitates the consideration of FLAs as serious threats to human health.

In the Philippines, the worsening problem with freshwater pollution causes detrimental effects to both its utility for human use, consumption, and aquaculture productivity [30]. The uncontrolled pollution of freshwaters systems in the country traced to indiscriminate dumping of industrial and domestic wastewater to rivers and other freshwater systems, the rapidly changing and expanding urban landscape causing soil runoff, and challenges brought upon by extreme weather conditions and climate change are contributory factors for the spread of waterborne protozoan pathogens [31,32]. FLA research in the Philippines is still in its infancy. To date, there are only two cases of fatal FLA-related encephalitis reported in the country [33,34] while 12 cases of non-fatal eye infections were documented [3,35,36]. This data, however, contradicts the high FLA isolation rate from environmental reservoirs frequented by people as documented by several recent studies [37–44]. The present study aimed to provide data on the current status of FLA epidemiology in the Republic of the Philippines, discuss the implications of environmental factors to human cases, and provide an inceptive roadmap for the progression of FLA investigation in the country.

## Methods

### Literature search strategy

Studies involving the biological and environmental isolation of *Naegleria* spp., *Acanthamoeba* spp., *Balamuthia* spp., *Sappinia* spp., and *Vermamoeba* spp., in the Republic of the Philippines, were searched systematically in PubMed and Google Scholar databases without years restriction. The search terms used to obtain the relevant studies were: '*Acanthamoeba*', '*Naegleria*', '*Balamuthia*', '*Sappinia*', '*Vermamoeba*', and 'Philippines'. To maximize the number of included studies and to prevent missing any relevant studies, the reference lists of included studies were searched for literature that can be included. This systematic review followed preferred reporting items for systematic reviews and meta-analyses (PRISMA) guidelines [45].

### Eligibility criteria

Titles of original research articles that reported FLA in the Philippines were screened. The inclusion criteria were: (1) studies with an abstract, (2) studies with full-text, and (3) studies written in the English or Filipino language. The exclusion criteria were: (1) Reviews and mini-reviews, (2) books and book chapters (3) method evaluation studies, and (4) intervention studies. Initial screening of retrieved records was performed by three authors (GDM, MFZH, and JP). Any disagreement with screening was resolved by consensus of three authors (GLM, EPM, and FRM). The final screening was performed by two authors (GDM and PK).

### Data extraction

Data such as year of study, study site, sources or types of samples, clinical manifestations, isolated FLA, genotypes, prevalence, methods used for identification, title, and references were extracted from the included studies. Data extraction was performed by five authors (GDM, MFZH, GLM, EPM, and JP) and was checked by three authors (GM, FRM, and PK) for accuracy.

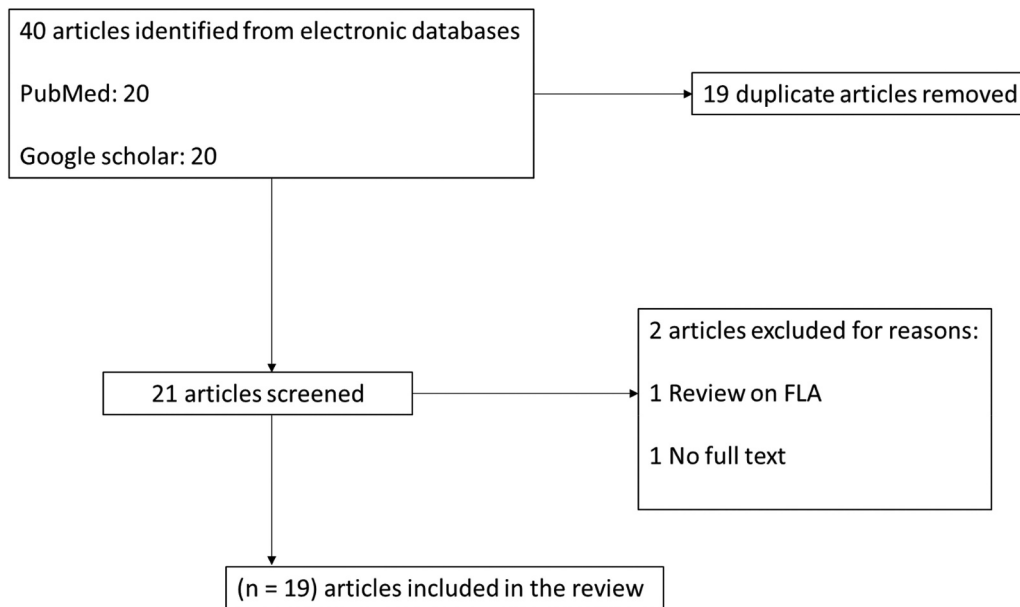
## Results

### Search results

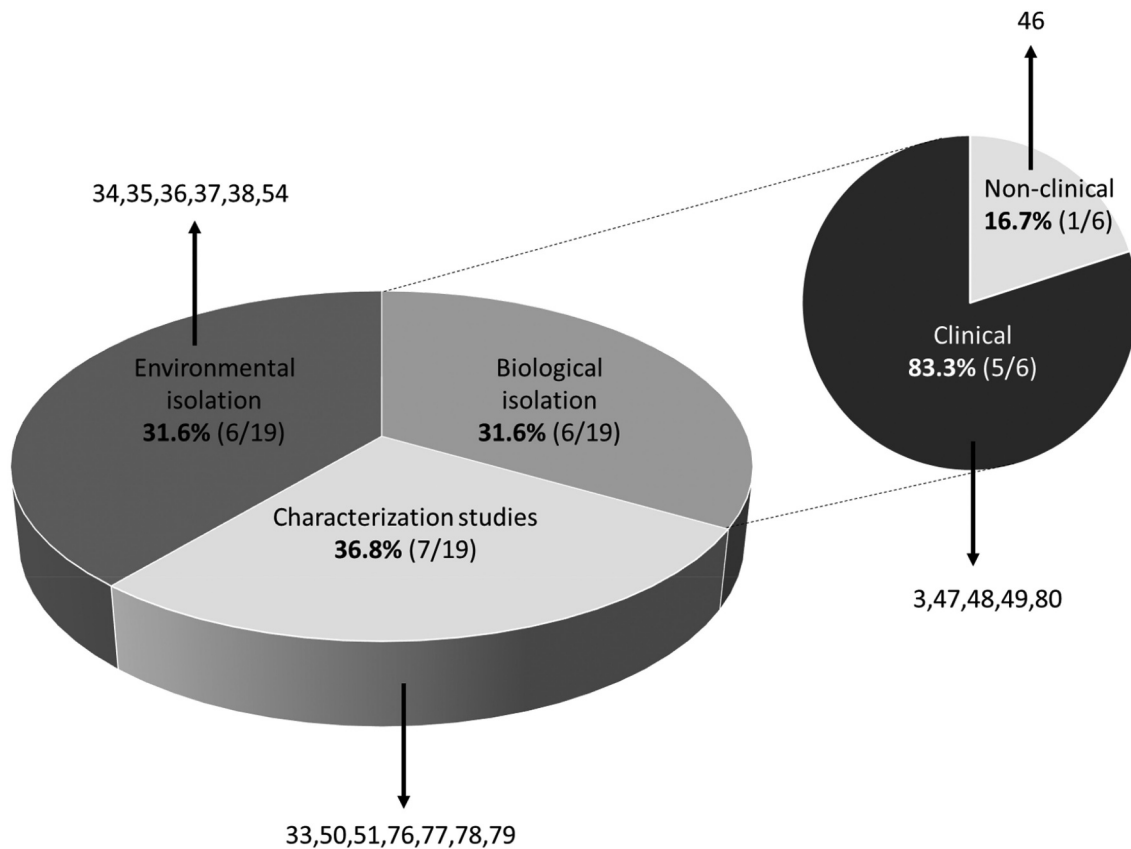
A total of 40 articles were retrieved from two databases (20 from PubMed and 20 from Google scholar). After the screening of abstracts and titles and retrieval of full-texts, 19 duplicate records were removed. Two out of 21 articles were excluded for the following reasons: Review paper (1), and no full text (1). Finally, a total of 19 articles were included in the present review (Figure 1).

### Characteristics of included articles

Among the included studies, 31.6% (6/19) focused on the isolation of FLA from environmental sources, such as soil, rivers, swimming pools, pond waters, lakes, and major water reservoirs in the Philippines. Biological samples composed 31.6% (6/19) of the included studies where 83.3% (5/6) reported FLA from clinical (human) samples while 16.7% (1/6) reported FLA from non-clinical samples, that is, fishes. FLA characterization comprised 36.8% (7/19) of the included studies and focused on the investigation of pathogenicity, thermo-tolerance, and chlorine resistance (Figure 2). Geographically, the majority of FLA detection was concentrated in the Island of Luzon where two genera of *Acanthamoeba* and *Naegleria* were identified from environmental sources, while only *Acanthamoeba* spp. was reported from the two other major Islands of Visayas and Mindanao (Figure 3 and Table 1). Only the genus of *Vermamoeba*, previously known as



**Figure 1.** Flow diagram of article search, screening, and selection.



**Figure 2.** Distribution of FLA studies in the Philippines according to study type. Numbers refer to included studies in the reference list.

*Hartmanella*, was reported from a non-clinical sample, that is, fish, while the genera of *Naegleria* and *Acanthamoeba* have been implicated in human infection cases in the country (Table 2).

The predominant *Acanthamoeba* genotype from environmental sources in descending order was: T4 and T5 (four reports each) > T15 (two reports) > T1, T3, T7, T9, T11, and T18 (one report each). *Naegleria*

**Table 1.** Environmental isolates of FLA in the Philippines based on molecular identification.

Island	Regions	Province/City	Sources	Species	Genotypes	References
Luzon	Region 1	Ilocos Sur	Pond water	<i>Acanthamoeba</i> spp.	T15	[40]
Luzon	Region 1	Ilocos Norte	Lake surface water	<i>Acanthamoeba</i> spp.	T1	[40]
Luzon	Region 3	Bulacan	Reservoir surface water	<i>Acanthamoeba</i> <i>graffini</i>	T3	[39]
Luzon	Region 3	Bulacan	Reservoir surface water	<i>Acanthamoeba</i> <i>castellanii</i>	T4	[39]
Luzon	Region 4a	Laguna	Lake surface water	<i>Acanthamoeba</i> spp.	T4	[42]
Luzon	Region 4a	Laguna	Lake surface water	<i>Acanthamoeba</i> spp.	T5	[42]
Luzon	Region 4a	Laguna	Lake surface water	<i>Acanthamoeba</i> spp.	T9	[42]
Luzon	Region 4a	Laguna	Lake surface water	<i>Acanthamoeba</i> spp.	T18	[42]
Luzon	NCR	Manila	Lake surface water	<i>N. australiensis</i>	NA	[44]
Luzon	Regions 3 & 4a	Cavite, Batangas, Pampanga	Lake surface water, Tap water, bottled water	<i>Naegleria</i> spp.	NA	[38]
Luzon	NCR, Region 4a	Manila, Cavite, Laguna	Soil and water	<i>N. philippinensis</i>	NA	[37]
Luzon	NCR, Regions 3 & 4a	Manila, Cavite, Batangas, Pampanga	River, Swimming pool, Pond water, Lake surface water	<i>Acanthamoeba</i> spp.	NM	[38]
Luzon	Region 5	Camarines Sur	Lake surface water	<i>Acanthamoeba</i> spp.	T4	[41]
Luzon	CAR	Ifugao	Reservoir surface water	<i>Acanthamoeba</i> spp.	T5	[39]
Luzon	CAR	Ifugao	Reservoir surface water	<i>Acanthamoeba</i> spp.	T7	[39]
Luzon	CAR	Ifugao	Reservoir surface water	<i>Acanthamoeba</i> spp.	T11	[39]
Visayas	Region 8	Leyte	Lake surface water	<i>Acanthamoeba</i> spp.	T5	[40]
Mindanao	Region 13	Agusan del Sur	Lake surface water	<i>Acanthamoeba</i> spp.	T4, T5, T15	[40]

NA Not applicable; NM Not mentioned; NCR National Capital Region; CAR Cordillera Administrative Region.

**Table 2.** FLA isolated from biological samples in the Philippines.

Year of isolation	FLAs	Genotypes	Sample	Clinical manifestations	Prevalence (%)	Methods	Suspected source	References
<b>Non-clinical (animal) sample</b>								
2017	<i>Vermamoeba vermiformis</i>	NA	Fish intestine	NA	100% (1/1)	Culture/Microscopy/PCR/Sequencing	Freshwater	[43]
<b>Clinical (human) samples</b>								
1984	<i>Naegleria philippinensis</i>	NA	CSF	PAM	100% (1/1)	Culture/Microscopy	Thermally polluted stream	[34]
1999	<i>Acanthamoeba</i> spp.	NM	CSF	AK	100% (1/1)	NM	NM	[33]
2009	<i>Acanthamoeba</i> spp.	NM	Corneal scraping	AK	100% (3/3)	Microscopy	NM	[35]
2014	<i>Acanthamoeba</i> spp.	T4	Corneal scraping	AK	100% (1/1)	PCR	Tap water	[36]
2014	<i>Acanthamoeba</i> spp.	T4 and T5	Nasal swab	None	4.44% (8/180)	PCR/Sequencing	Garbage, soil, and dust	[3]

NA not applicable; NM not mentioned; PAM primary amoebic encephalitis; AK *Acanthamoeba* Keratitis; PCR polymerase chain reaction.

was reported in three studies where *Naegleria* spp., *N. australiensis*, and *N. philippinensis* were isolated from water samples while *N. philippinensis* was also isolated from soil and water samples.

For clinical (human) samples, two genera of FLA, *Acanthamoeba*, and *Naegleria* were identified with *Acanthamoeba* having a total of 13 cases while *Naegleria* has only one case reported. The point sources of clinical (human) isolates could not be mapped as the detection of FLA, that is, *Acanthamoeba* and *Naegleria* were all performed in the city of Manila with the suspected source of FLA either not mentioned or being speculative only (Table 2). The predominant genotype of *Acanthamoeba* isolated from human samples was T4 (two reports) followed by T5 (one report) while *N. philippinensis* was only reported once from a PAM patient (Table 2).

No Philippine reports on *Balamuthia* and *Sappinia* were retrieved.

## Discussion

### Distribution and diversity of FLA in the Philippines

Isolation and identification of FLA in different environmental sources, especially those considered pathogenic, at present, is rare in the Philippines where matrices, such as soil, water sediments, biofilms, dust, and air, require exploration. It is hypothesized by the authors that the archipelagic profile and abundance of freshwater systems in the Philippines support a wide spectrum of FLA biodiversity that remains unexplored. This warrants the promotion of environmental surveys for FLA biodiversity in the country. Although there are defined surveillance studies conducted for other parasites like soil-transmitted helminths [46,47], there appears to be none for FLA despite the molecular identification of *Acanthamoeba*, *Naegleria*, and *Vermamoeba* from the soil, freshwater, and fishes in the country [37–43]. Surveillance studies are important not only in identifying the existence of pathogenic

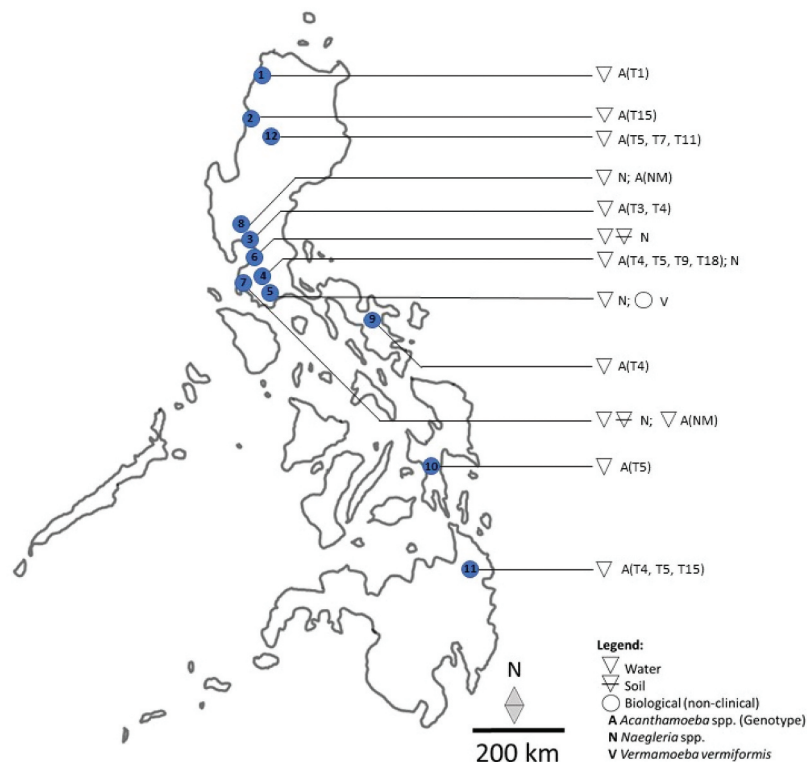
species and genotypes but also in defining multiple transmission routes involving the environment, animals, and humans. This in turn will lead to the formulation of strategies in the mitigation and management of FLA infections. National surveillance studies for FLA exist in countries like the United States, which enables the health-care sector to monitor outbreaks [48]. In Luzon Island, water reservoirs that are vital freshwater sources have demonstrated the presence of *Acanthamoeba* genotypes T3, T4, T5, T7, and T11 [39] while *Naegleria* spp. were reported from environmental sources as well as bottled water [37,38,44]. In the Visayas Island, Lakes Danao, and Bito in Ormoc, Leyte were positive with *Acanthamoeba* genotype T5 while three genotypes T4, T5, and T15 were isolated in Lake Mainit in the Island of Mindanao [40].

### FLA infections in the Philippines

Despite the reported diversity of FLA and pathogenic genotypes in various environmental sources in the country (Figure 3), reports on morbidity and/or mortality cases of FLA-related infections remain largely unknown. *Acanthamoeba* keratitis (AK) was first reported in the country in 1992 [33]. Between 2002 and 2004, three more cases of AK were reported [35]. In 2009, a 76-years-old male reported pain, redness, and visual impairment of

the right eye where molecular testing confirmed *Acanthamoeba* genotype T4 [36]. It is important to note that all these AK cases involved non-contact lens wearers and that the latter case used tap water to cleanse the face before the onset of symptoms. Surprisingly, in the study of Cruz and Rivera (2014), 4.4% (8/180) of nasal swabs from asymptomatic volunteers were positive with *Acanthamoeba* spp. T4 and T5 [3]. The proposed transmission route was obviously from the chronic exposure of the volunteers to garbage, soil, and dust as part of their everyday activity. In this case, it is interesting to speculate on the potential health outcomes in the event of depression or suppression of the immune system of the *Acanthamoeba*-positive volunteers. Also, it opens up perspectives into the health hazards on FLA-related infections faced by high-exposure groups of laborers working in unsanitary conditions and environments. Also, the predominance of *Acanthamoeba* genotype T4 and T5 in local human cases was potentially influenced by the propensity of the same genotypes reported in the highest frequencies in local environmental sources.

To date, there is only one report of *Naegleria* causing PAM infection in the Philippines [34]. This 1984 isolate was later identified as a new species of *Naegleria* and was given the name *N. philippinensis*



**Figure 3.** Geographical distribution of FLA from environmental and biological (non-clinical) samples in the Philippines. 1 Ilocos Norte; 2 Ilocos Sur; 3 Bulacan; 4 Laguna; 5 Batangas; 6 Metro Manila; 7 Cavite; 8 Pampanga; 9 Camarines Sur; 10 Leyte; 11 Agusan del Sur; 12 Cordillera Autonomous Region; A *Acanthamoeba*; (Genotype); N *Naegleria*; V *Vermamoeba*; NM genotype not mentioned. Note that FLA isolation in humans could not be mapped as the point sources were either not mentioned or speculative only.



after a series of molecular characterization and pathogenicity assays and was suspected to have been contracted from a thermally polluted stream [49]. Although *N. philippinensis* was not reported from environmental sources in the Philippines, the isolation of *N. australiensis* from a Lake surface water in the National Capital Region (NCR) poses a health threat to the general public [44]. In addition, the isolation of *Vermamoeba vermiformis* from a freshwater fish (*Oreochromis niloticus*) in a volcanic lake (Lake Taal) in Luzon Island [43] strengthened the perspective of the role of fishes as biological reservoirs for FLAs [50–53].

### Methods of isolation and identification of FLA

From the first local isolation and characterization of *Naegleria* and *Acanthamoeba* from human samples [33,34], the use of light microscopy in the detection and description of FLA has proven valuable in aiding clinicians in the course of disease management. However, with the advent of new techniques and the introduction of molecular testing, there was an improvement in the specificity and accuracy of detection and identification of FLAs [3,36].

In the case of point source isolation from aquatic systems, modifications in sample collection have been introduced for the convenient, fast, and cost-effective detection of waterborne protozoan pathogens [54,55]. From obtaining at least 10 L water samples [37,38], the introduction of low-volume water sampling (50 mL for microscopy and 1000 mL for molecular assay) and manual filtration using a 1.2 µm pore-size filter has proven reliable for detecting waterborne protozoan pathogens from surface water, sediments, and substrate-associated biofilms [41,42,44,56,57].

Previous identification of FLAs was solely based on phenotypic characteristics with the aid of light microscopy and the use of an enflagellation test to differentiate *Naegleria* from *Acanthamoeba* [33,34]. However, some studies have demonstrated the limitations of the enflagellation test [58]. The use of polymerase-chain reaction (PCR) for the detection and identification of FLAs enabled researchers to accurately identify interspecies differences [39–42]. Also, *in-vitro* analysis such as osmotolerance and thermotolerance assay, encystment rate, scanning electron microscopy, and molecular techniques in combination helped to further characterize the species, genotype, and pathogenicity of isolates [40]. Regardless of the identification technique, culture was key in the successful isolation of FLAs from environmental and biological samples.

The continuous evolution of FLAs through their adaptability to different environmental conditions warrants its exploration in a wide variety of environmental, infrastructural, biological, and clinical regimes, as with

the recent isolation of *Acanthamoeba* from seawater and hospital ward dust [59] and from the urinary tract of humans [60]. These are suggestive of the role of FLAs as latent carriers for resistant bacteria and biofilm formation as well as the introduction of FLA stages during invasive and operational procedures.

### Current status of FLA study in the Philippines

FLA-related research in the Philippines is still in its inceptive stage. To date, only a fraction of the 101 lakes scattered across the country [61] has been explored for FLA biodiversity. Although the first nationwide study on FLA diversity was successful in defining previously unreported species and genotypes [40], more research is necessary to further establish its biodiversity from a wide range of environmental, infrastructural, and biological regimes.

The presence and proliferation of FLA in aquatic systems may, in part, be due to rapid urbanization. The increased disturbance caused by informal human settlements near lakes and other freshwater bodies thereby increases domestic waste pollution, soil runoff, human and animal waste pollution, and thermal pollution from industries that dump wastes directly to freshwater bodies [44,62]. Also, the increasing temperature of waterbodies due to climate change potentially enhances the thermotolerance of the FLA fauna [40]. Further, meteorological events like flooding facilitate the widespread dispersal of waterborne protozoan pathogens including FLA from terrestrial to aquatic environments and back [55].

Despite the environmental ubiquity of FLA, FLA-related infections in the Philippines remain low. In the last 36 years, only 13 cases of *Acanthamoeba* and 1 case of *Naegleria* infection were documented in the country, two of which led to fatal outcomes [33,34]. The current hygienic status of public hospitals and health-care centers in the country is appalling [63]. The poor distribution and accessibility of health-care facilities especially in rural settings, the general lack of knowledge on FLA, the limited use of existent international protocols for identification of FLA-related infections, and resource limitations may be contributory factors for the rarity of reports on FLA-related infections or the lack of it in the Philippines. This was observed in a five-year report (2016–2020) of the Epidemiology Bureau on the distribution of Acute Meningitis-Encephalitis Syndrome (AMES) cases in the Philippines, which shows a very high number of unclassified encephalitis cases from different regions of the country (see supplemental data). Similarly, this was observed in the United States where the high occurrence of unspecified causes of meningitis was observed [64]. It is also important to consider the possibility of an acquired immunity or tolerance among subpopulations to certain strains of potentially

pathogenic FLAs [3]. Some studies have suggested the presence of mammalian antibodies against AK causing *Acanthamoeba* genotypes and those that cause fatal brain infections [65,66]. Although there is no direct evidence on this, it should be taken into perspective. *In-vitro* pathogenicity assays demonstrated variations in FLA pathogenicity and suggest that pathogenic and nonpathogenic genotypes exist within previously established pathogenic groups [40,67]. These factors may rationalize the uneven proportion of morbidity and/or mortality cases to high isolation rates of FLAs from environmental sources.

The recent isolation of *Vermamoeba vermiformis* from the intestine of an edible freshwater fish (*O. niloticus*) opened a new direction in FLA studies in the Philippines [43]. Although no evidence of pathogenicity to its fish host has been documented, this FLA is capable of hosting endocytobionts/endosymbionts within its cytoplasm, which may induce pathogenicity to humans [68,69]. This is suggestive of the role of fishes as vectors or biological reservoirs for FLAs to proliferate and persist in aquatic environments and potentially infect fishermen, fish handlers, and those consuming raw fish, in particular, small fishes that are not eviscerated and eaten whole. *Acanthamoeba* and *Naegleria* have demonstrated propensity in aquatic sources in the Philippines. With evidence of the pathogenicity of these isolates [40], there is a potential risk of infection for those in direct contact with contaminated water sources. The proliferation of these pathogenic FLAs in various environmental settings can be mitigated by focusing on the implementation of control efforts on different factors mentioned previously.

### Challenges and inceptive roadmap for FLA research in the Philippines

The Philippines is an archipelago of more than 7,100 Islands rich in freshwater ecosystems hypothesized to be teeming with FLA biodiversity and potentially pathogenic species and genotypes. However, a case definition for FLA-related or suspected infection is not included in the country's Acute Meningitis-Encephalitis Syndrome (AMES) surveillance program of the Epidemiology Bureau Public Health Surveillance Division of the Department of Health. Although a nationwide study on FLA distribution was recently conducted in the Philippines [40], a plethora of study sites and biological sources remains unexplored.

A 2021 Thailand study surveyed public freshwater sources across 28 provinces and offered a treasure trove of information in the development of FLA study in a country. It demonstrated the value of defining FLA distribution and biodiversity. The survey of 100 sources demonstrated the isolation of nine previously reported (T2/6, T3-T5, T9, T11, T12, and T18) and one novel genotype (T23) proposed to be

*Acanthamoeba bangkokensis*. Also, a study of this scale and depth established the following: (1) the predominant species or genotype, (2) the reproductive success of FLA in diverse ecological niche, (3) perspectives on cryptic subtypes within defined genotypes, (4) potential discovery of novel species/genotypes, (5) evolution, (6) data on genetic and morphological characteristics useful for identification, and (7) the establishment of a library of sequences and isolates for in-vivo and in-vitro studies [70]. Such information is valuable for widespread information dissemination to the public, scientific, and medical community.

Success in the progression of FLA study in the Philippines entails the exploration of a wide variety of sources, which aside from the natural environment, also includes infrastructure/gray structures [59], unexplored or less explored sample matrices like sediments and biofilms [71,72], and anatomic sites/clinical regimes not previously thought to be colonized by FLAs [60].

Collaborations with local government units as well as members of the local communities for information dissemination on safety protocols against contracting FLA infections like wearing goggles and nose clips should be strongly advocated among fishermen and other stakeholders. This may also help in building a local industry on locally made goggles and nose clips using recyclable materials that will not only aid in health promotion but a sustainable livelihood among locals. Likewise, establishing the distribution and identity of FLAs and its possible endosymbionts [73] in various sources will help to form perspectives on potential outcomes for the general human population as well as its subpopulations, primarily, immunocompromised individuals who are at the highest risk of developing progressive and fatal infections [74].

The lack of standardized management and effective drug for FLA-related human infections necessitates drug discovery. The Philippines is rich in natural products [75] that may show potential anti-protozoan activity against FLA-related morbidity. From a public health perspective, a multi/trans-disciplinary approach should be employed in the mitigation of FLA human infections, particularly in water-related occupations, tourism, and recreational sectors.

To the best of our knowledge, no report in FLA-related infections was ever included in the AMES national case reports of the DOH (see supplemental data). Also, no training and funding has been allocated to the identification of FLA in the Food and Water-Borne Disease Strategic plan for 2019–2023 in the country. This is strong evidence that FLA-related pathophysiology is either still relatively unknown or neglected and is a gaping hole in the health research agenda in the Philippines. The outcomes

of the present review necessitate that a case definition for FLA-related infections be included in the country's AMES surveillance program. Finally, studies on the potential of FLA to become a bioindicator of water pollution/contamination have been proposed by recent studies in selected lakes in the country [76 77 78 79 80 81].

### Conclusions

FLA studies in the Philippines are fragmented and are still in their inceptive stage. Gathered literature highly suggests a severe lack of FLA research in the country. These gaps, however, can be translated to promising FLA-related studies following perspectives for a national FLA research road map. Despite the ubiquity of FLA in environmental sources in the country, documented human cases are rarely reported. The rarity of cases is most likely a result of underreporting due to the absence of a national case definition, lack of health facility, and/or the lack of diagnostic capacity to distinguish FLA infections from bacterial and viral-induced pathologies. FLA-related infections are a public health threat in the Philippines and demand further studies in identifying its epidemiologic aspect in both clinical cases and environmental sources to define the morbidity/mortality rates of this neglected parasitic infection.

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