



Surveys of Canine Filarioids In Nigeria: The Path Travelled and the Way Forward

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SUMMARY

Filarioid worms infecting dogs have recently received increased attention globally because of their zoonotic potential. In Africa and, particularly, in Nigeria, however, where there is preponderance of the risk factors for vector-borne diseases transmission, there are few reports of the disease in the canid and felid definitive hosts, the wild/domestic reservoirs and humans. Thus, the epidemiology of the disease in Nigeria remains sketchy and needs to be investigated. A retrospective analysis of reported canine filarioids in Nigeria was undertaken with the view to highlight what has been done and reported, existing gaps in knowledge, what needs to be done to bridge the gap and possibly how it could be done. Thirteen published works on canine filarioids using classical laboratory methods in Nigeria, reported the finding of *Dirofilaria immitis* (0.4–15.1%), *Dirofilaria repens* (0.1–9.4%), *Acanthocheilonema reconditum* (0.4–9.2%) and a case of *A. dracunculoides* in an unspecified dog population. In most instances, the species identification of the filarioids reported was not conclusive due to limitation of the diagnostic methods employed. No human infection due to any zoonotic canine filarioid has been reported in Nigeria. Suggestions for prospective filarioids research in Nigeria were made.

Keywords: Filarioids, dog, zoonosis, diagnosis, Nigeria.

INTRODUCTION

Canine arthropod-borne filarioids include nematodes of the superfamily Filarioidea

which are transmitted by arthropods such as mosquitoes, fleas, flies, lice or ticks depending on the genus of the filarioid

involved (Nelson *et al.*, 1962; Farnell and Faulkner, 1978; Anyanwu *et al.*, 2000; Otranto *et al.*, 2013). Canine filarioids are widespread in the tropical, sub-tropical and temperate zones (Pampiglione and Rivasi, 2000; Otranto and Deplazes 2019). Over the years, human activities and climate change have increased the geographical range of filarioid worms and consequently the risk of infection for both animals and humans. Likewise, advances in diagnostic methods, greater attention to animal welfare and improved social and economic conditions in many countries have contributed to an increase in pet owners' awareness of potentially life-threatening conditions (Colwell *et al.*, 2011). Therefore, intensified research efforts globally have identified several filarioids such as; *Dirofilaria* spp., *Acanthocheilonema* spp., *Brugia* spp. and *Onchocerca lupi* among the most important species affecting dogs (Eberhard, 1980; Orihel and Eberhard, 1998; McCall *et al.*, 2008; Pampiglione *et al.*, 2009; Otranto *et al.*, 2011; 2017).

However, in most parts of Africa and in other developing regions of the world where the climatic conditions are favorable for the transmission of filarioid worms, there is paucity of information on canine and/or human filariosis. This may be attributed to inadequate research funding and poor diagnostic services (Orihel and Eberhard 1998; Simón *et al.*, 2017). In Nigeria, Schillhorn van Veen, (1974) documented the earliest evidence of canine filarioid infections following a survey of dogs in the Zaria area. From the earliest report where *D. repens* was identified, other survey studies

and case reports have incriminated other species of filarioid worms infecting dogs in Nigeria such as *D. immitis* and *A. reconditum*. Going by the number of publications it will be surmised that remarkable progress has been made in the area of canine filarioids research in Nigeria (Schillhorn van Veen, 1974; Oduye and Dipeolu, 1976; Idowu *et al.*, 1977; Bobade *et al.*, 1981; Kamalu, 1986, 1991; Uche and Odunze 1988; Anyanwu *et al.*, 1996, 2000; Abah, 2015; Obaje and Abel-Danjuma, 2016; Ugochuckwu *et al.*, 2016). But in the authors' opinion there still exist gaps in knowledge of the basic epidemiology of the disease in the country, which could be extrapolated to the rest of the African continent, that need to be addressed. The aim of this paper is to review the findings from previous publications in line with established guidelines for diagnosis of canine filarioids and to highlight existing knowledge gaps and recommend possible areas for future research.

Canine Filarioid Species Previously Described In Nigeria

Identification of four canine filarioid species (*D. immitis*, *D. repens*, *A. reconditum* and *A. dracunculoides*) have been documented so far in Nigeria. They primarily affect different species of carnivores, mainly canids, and are transmitted by haematophagous arthropods: *Dirofilaria* spp. by mosquitoes, *A. reconditum* by fleas and lice and *A. dracunculoides* by fleas and ticks (Magnis *et al.*, 2013) (Fig. 1). Adult worms of *D. immitis* are mainly located in the pulmonary

artery and right ventricle of canine hearts (Simón *et al.*, 2012). *Dirofilaria repens* and *A. reconditum* are habitual parasites of the subcutaneous tissue (Simón *et al.*, 2012; Engelmann *et al.*, 2018), whereas the predilection sites of the adult worms of *A. dracunculoides* are the abdominal and thoracic cavities (Fig. 1). Adult female worms release microfilariae from these locations into the bloodstream of the final hosts. Their morphological characteristics could be of some importance in diagnosis and are summarized in Table I.

Regarding pathology, *Dirofilaria immitis* is responsible for a chronic inflammatory vascular disease, which can trigger pulmonary hypertension and congestive

heart failure, and *D. repens* can cause some dermatological problems (McCall *et al.*, 2008; Simón *et al.*, 2012). Although the species of the genus *Acantocheilonema*, were previously regarded as non-pathogenic to dogs (Nelson, 1966), there are cases of pruritic dermatosis and focal alopecia in dogs attributed to the action of *A. reconditum* microfilariae when it is present in large numbers (Bobade *et al.*, 1981). Dermal clinical signs and lesions such as pruritus, alopecia, erythema, skin ulcers as well as other clinical signs such as ataxia, incoordination, cachexia, cyanosis, ascitis and pleural effusion have been occasionally associated with the *A. dracunculoides* infection (Piercy, 1951; Bolio-Montes *et al.*, 2002).

TABLE I. Morphological characteristics of the canine filarioid species previously described in Nigeria.

	<i>D. immitis</i>	<i>D. repens</i>	<i>A. reconditum</i>	<i>A. dracunculoides</i>	References
Cephalic end	Conical	Conical with 2-3 nuclei in the head space	Long clear head space, cephalic hook	Clear head space, no cephalic hood	Taylor, 1960; Marconcini <i>et al.</i> , 1996; Schwan & Durand, 2002
Caudal extremity	Acute with nuclear column extending to the end of the body	Acute with nuclear column not extending to the end of the body	Attenuated tail, free of nuclei	Short attenuated tail, free of nuclei, bluntly ending	Taylor, 1960; Kamalu, 1986; Marconcini <i>et al.</i> , 1996
Shape of the unfixed and unstained tail	Straight	Umbrella handle	Hook	Straight	Nelson, 1962; Bobade <i>et al.</i> , 1981; Marconcini <i>et al.</i> , 1996
Size(wxI)	234-260µm and 4-6µm	300-369µm and 7-8µm	225-282µm	240-260 µm and 5-6 µm	Leger, 1911; Bouin, 1921; Marconcini <i>et al.</i> , 1996

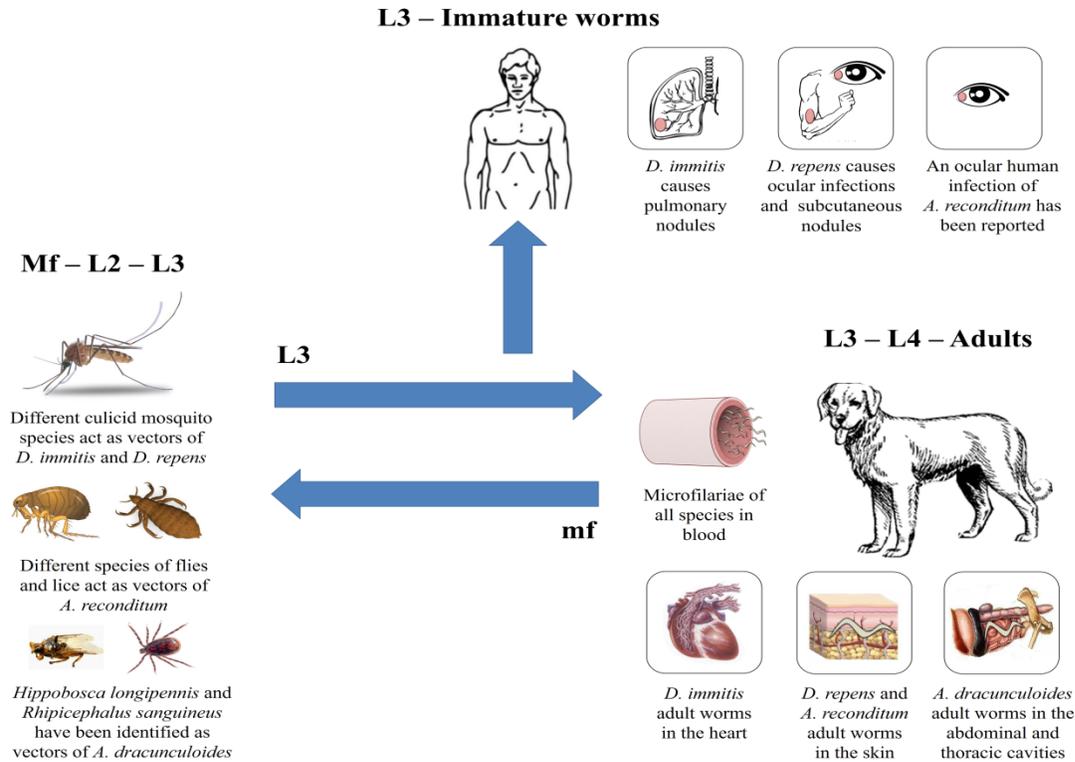


Fig 1 Schematic representation of the biological life cycles of the canine filarioid worms reported in Nigeria

Zoonotic Implications And Public Health

Some of the canine filarioids reported in Nigeria have the potential to infect humans and remain important from a public health perspective (Fig. 1) (Irwin and Jefferies, 2004; Sabu *et al.*, 2005). *Dirofilaria immitis* human infections are usually caused by immature worms reaching a branch of the human pulmonary artery, triggering an inflammatory response that destroys the worms and occasionally resulting in pulmonary nodules (Pampiglione and Rivasi 2001; Simón *et al.*, 2012). There are over 450 reported cases of human *D. immitis* pulmonary infections worldwide (Simón *et al.*, 2017). In most cases the infections were

asymptomatic, showing typical ‘coin lesions’ on chest radiographs which are often mistakenly removed as neoplasms (Simón *et al.*, 2005), thus reducing the overall tally of the disease in humans.

Dirofilaria repens causes sub-conjunctival infections and subcutaneous nodules often involving organs such as lungs, eyes, male genitals and female breast (Pampiglione and Rivasi, 2000; Otranto and Eberhard, 2011) and it often leads to misdiagnosis of malignant tumors in endemic areas (Simón *et al.*, 2005). There are over 4490 reported human cases due to *D. repens* infection globally (Simón *et al.*, 2017). Therefore, parasitosis due to *D. repens* is considered as

an emerging zoonosis in many geographical areas.

Regarding human *Acanthocheilonema* infections, a case with *A. reconditum* involving the eye of the patient has been reported in Australia (Huynh *et al.*, 2001). Surprisingly, of all the reported human infections attributed to canine zoonotic filarioid worms, there are very few cases (less than 20) from Africa (Simón *et al.*, 2017). To date only 15 human cases of *D. repens* infections, have been reported in Tunisia and one each from Tanzania and South Africa (Pampiglione and Rivasi, 2007; Saeid *et al.*, 2011; Moodley *et al.*, 2015). So far there is no documented report of human infection due to filarioids of canine or feline origin in Nigeria. This could be attributed to lack of awareness and inadequate diagnostic facilities.

Diagnosis of canine filarioids

Several diagnostic methods aimed at the detection of the adult worms, circulating microfilariae or parasite antigen or DNA of varying sensitivities and limitations have been developed over the years (Mar *et al.*, 2002; Latrofa *et al.*, 2012a, b). Diagnostic challenge of canine filarioids stems from the variation in the predilection site of the adult worms and larvae (microfilariae) as well as the exhibition of periodicity by microfilariae of certain species (Latrofa *et al.*, 2012a). Furthermore, dogs with pre-patent or occult heartworm infections are usually amicrofilariaemic while those on macrolide prophylactic therapy may be amicrofilariaemic, hence confounding the diagnosis (Nelson *et al.*, 2005). In the case of *D. immitis*, although it has been

historically reported in dogs in some countries of Africa, including Nigeria (Genchi *et al.*, 2001), there is a great deal of confusion over its identification and prevalence. The distribution of the disease in Africa is not well known due to the paucity of epidemiological studies, the lack of methodological details regarding the assays employed for those studies, and the large variety of filaria species endemic to this continent (Simón *et al.*, 2012). Thus, there is the need to accurately identify any microfilaria detected in the blood of dogs before making a diagnosis. However, as their size ranges overlap (Table I), it is unreliable to establish a diagnosis on this criterion alone. In this sense, molecular approaches based on PCR methods have recently allowed researchers to make more accurate identifications in the canine population of some countries in Africa, such as Cape Verde (Pereira *et al.*, 2013; Marcos *et al.*, 2017), Tanzania (Mukendi *et al.*, 2016), Tunisia (Rjeibi *et al.*, 2017) and Algeria (Tahir *et al.*, 2017). Therefore, there is the need to combine two or more diagnostic methods before arriving at a confirmatory diagnosis.

Appraisal Of Canine Filarioids Research In Nigeria

Several research groups have reported the detection of filarioid worms of dogs from different parts of Nigeria either as case reports or survey studies (Fig. 2). In all, 13 reports of canine filarioids using classical methods were published in the literature (Table II).

The earliest report of filarioid infections in dogs in Nigeria dates back to 1974

(Schillhorn van Veen, 1974). In two separate survey studies using morphologic and morphometric criteria for identification of microfilariae of dogs in Zaria, northwest Nigeria, *D. repens* (300–369 μm) and *A. reconditum* (234–264 μm) were detected in 9.4% and 9.2%, respectively, of the study population (Schillhorn van Veen, 1974; Schillhorn van Veen and Blotkamp, 1975). Although the length of microfilariae cannot be relied upon solely for a definitive diagnosis (Ciocan *et al.*, 2010), the measurements provided by the authors falls within the range of the species identified based on data available in the literature (Taylor, 1960). Similarly, the only report of *A. dracunculoides* in dogs in Nigeria was made in Zaria in 1974. Thus, according to Schillhorn van Veen, (1974) dogs in Zaria, Nigeria are not infected with *D. immitis*. However, disparate from the view of Schillhorn van Veen, (1974) on *D. immitis* infection in dogs in Zaria, later studies reported *D. immitis* –like and *D. immitis* in 2.5% and 15%, respectively, in dogs in the same study area (Anyanwu *et al.*, 1996; Abah, 2015). In other parts of Nigeria, 0.4% of 488 dogs and a single case report involving a three-year-old male Doberman appear to be the only reports of *D. immitis* infection in Ibadan Southwest Nigeria (Idowu *et al.*, 1977; Ajadi *et al.*, 2011), suggesting that *D. immitis* infection of dogs is rare in Southwestern Nigeria (Fig. 2). Recently, for the first time in Southeastern and Northcentral regions of Nigeria, *D. immitis* infection was reported in 3.4% of 119 and 1.6% of 186 dogs, respectively examined by using wet mount and buffy

coat techniques (Ugochukwu *et al.*, 2016; Obaje and Danjuma, 2016).

Dirofilaria repens was reported in five studies with prevalence ranging from 0.1% to 9.2% (Schillhorn van Veen, 1974; Uche and Odunze, 1988; Kamalu, 1986, 1991; Anyanwu *et al.*, 1996). Other filarioid species reported within this period were *A. reconditum* and *A. dracunculoides* (Schillhorn van Veen, 1974; Schillhorn van Veen and Blotkamp, 1975; Idowu *et al.*, 1977; Bobade *et al.*, 1981; Uche and Odunze, 1988). Some of the studies could not conclude on the identity of the microfilariae, thus they reported their findings as unidentified microfilariae. Overall, four species of filarioid worms: *D. immitis*, *D. repens*, *A. reconditum* and *A. dracunculoides* have been reported in dogs in Nigeria to date (Table II). What constituted the reported unidentified microfilariae in the various studies is a subject for further investigation.

Identification of canine filarioid species in all the reported studies in Nigeria relied on the classical diagnostic methods such as the wet mount, Knott's modified method, Haematocrit centrifuge technique (HCT) and thin smear, singly or in combination for the identification of adult worms or circulating microfilariae. These diagnostic methods have low sensitivity and are therefore potentially misleading. The modified Knott's technique is sensitive but requires highly experienced personnel to interpret the result. Even at that, it is difficult to detect multiple infections with more than one species of filarioid worms or

accurately differentiate between the microfilariae of the various species, thus leading to false results (Scoles and Kambhampati, 1995; Rishniw *et al.*, 2006). These limitations notwithstanding, available data on canine filarioids in Nigeria appear to reveal a change in disease pattern over the years. Notably, there appear to be an increase in disease prevalence as well as a

switch in the prevalence of *D. repens* and *D. immitis*. These changes deserve elucidation in order to ascertain the true status of the disease in the canine population in Nigeria. Our approach is to undertake a retrospective analysis of previous literature reporting canine filarioids in Nigeria in line with current scientific

TABLE II: Reports of canine Filarioid infections in Nigeria, 1974-2017

s/no	Period of sampling	Study area (Town-region of Nigeria)	Sample size	Reported prevalence (%)					Method(s) used	Reference	Number of citations (as at December 2017)
				<i>D. immitis</i>	<i>D. repens</i>	<i>A. reconditum</i>	<i>A. dracunculoides</i>	Unidentified microfilariae			
1	1974	Zaria-Northwest	NA	0	9.4	0	yes	0	Morphology of adult worm	Schillhorn van Veen, 1974	8
2	1975	Zaria-Northwest	369	0	0	9.2		yes	concentration	Schillhorn van Veen & Blotkamp, 1975.	4
3	1974-1975	Ibadan-Southwest	500	0	0	0	0	yes	Blood smear	Oduye & Dipeolu, 1976	30
4	1972-1975	Ibadan-Southwest	488	0.4	0	0.4	0	0	Wet mount, Modified Knott's technique	Idowu <i>et al.</i> , 1977	9
5	1977-79	Nsukka Southeast	3541	0	0.1	0	0	0	Modified Knott's technique	Kamalu, 1986	10
6	1981	Ibadan - Southwest	Case report	-		Yes	0	-	Thick smear, Modified Knott's technique	Bobade <i>et al.</i> , 1981	2
7	1985	Nsukka-Southeast	200	0	1.5	2.5	0	0	Wet mount, Buffy coat Modified Knott's technique, autopsy	Uche & Odunze, 1988	2
8	1991	Nsukka Southeast	Case report	0	yes	0	0	0	Morphology of microfilariae and adult worm	Kamalu, 1991	22
9	1995-1996	Zaria-Northwest	NA	2.5	8.9	0	0	4.4	Blood smear	Anyanwu <i>et al.</i> , 1996	4
10	2011	Ibadan-Southwest	1-Case report	Yes	0	0	0	0	Blood smear	Ajadi <i>et al.</i> , 2011	0
11	2012	Zaria-Northwest	317	15.1	0	0	0	0	Modified Knott's technique	Abah, 2015	0
12	2015-2016	Makurdi-Northcentral	186	1.6	0	0	0	0.5	Wet mount, Buffy coat Modified Knott's technique	Ogbaje & Danjuma, 2016	0
13	2016	Nsukka-Southeast	119	3.4	0	0	0	0	Wet mount, buffy coat, autopsy	Ugochuckwu <i>et al.</i> , 2016	0

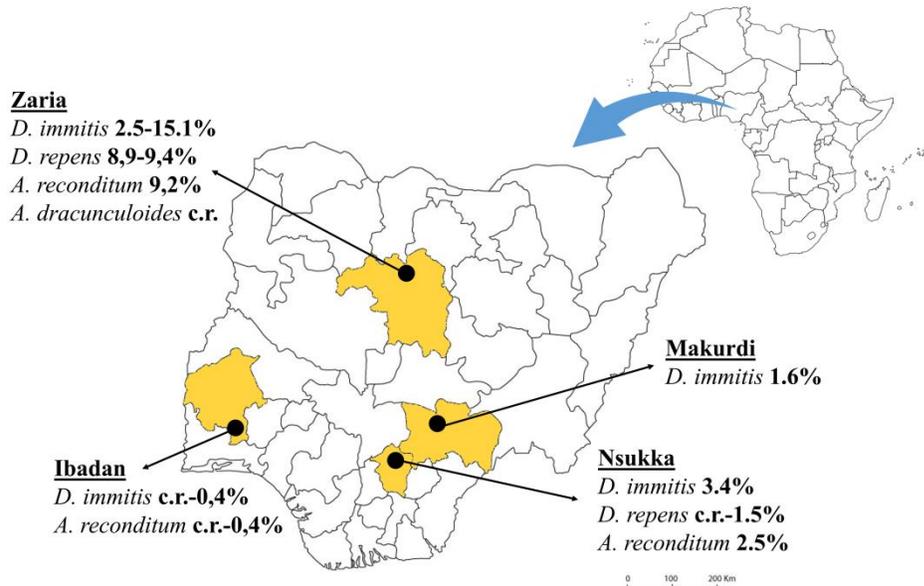


Fig 2 Map of Nigeria, West Africa showing reported prevalence of canine filarioids (c.r.= case report)

knowledge and available information on canine filarioids globally with the view to determining predominant species reported in this country and their geographical spread. Nelson, (1966) observed earlier that it was common in veterinary practice to assume that dogs with microfilariae in their blood were infected with *D. immitis* which has led to great deal of confusion with other harmless species. Thus, researchers should guard against the assumption that every motile, thread-like slender organism or every microfilaria seen in the blood of dogs is indeed *D. immitis*.

Knowledge Gaps

From the forgoing, there is no doubt that there have been modest efforts from the Nigerian research community to determine the species and prevalence of canine filarioids over the last four decades. However, the epidemiology of the disease still remains largely unknown due to some intrinsic and extrinsic limiting factors in the Nigerian research landscape. This may be attributed to poor funding of research, improper research design, diagnostic approach, lack of specialized facilities and trained personnel. Drawing hasty conclusions may partly be responsible for the present state of knowledge.

Unfortunately, this has led to a quantity of publication of preliminary unauthenticated results serving as “dangerous references” thereby misleading the international research community (Otranto, 2015). Therefore, researchers should endeavor to exercise restraint and guard against releasing preliminary unverified results until they are conclusively verified in line with set guidelines before releasing such to the larger research community (Uilenberg *et al.*, 2018).

It should be borne in mind that accurate diagnosis is pivotal to the treatment and prevention of parasite infections. Therefore, diagnostic techniques should be chosen on the basis of what information is needed as well as their sensitivity and specificity (Otranto, 2015). Thus far, no study of canine filarioids in Nigeria had employed any molecular method or other advanced techniques in addition to the classical methods, in order to accurately speciate the parasite and authenticate the results.

Reading through the reports, it was observed that the earlier authors were more circumspect and sought expert opinion where possible before arriving at final diagnosis. Sometimes they left their diagnosis at the genera level, or simply refer to the microfilariae as ‘unidentified’. Not so the latter authors who gave definitive speciation without consideration to the limitations of the diagnostic approaches used or to set guidelines. Equally, the earlier reports were made when molecular diagnostic methods were not available or at their infancy hence the authors relied on the

readily available techniques then. It would seem fortunate for the latter authors to take advantage of the current advances in molecular techniques and availability of avenues for collaborations in order to validate their findings.

The Way Forward

As it stands there exists a lacuna and further work is needed to determine the real identities and true prevalence of canine filarioids present in Nigeria. Future studies should take cognizance of the established guideline for the laboratory diagnosis of canine filarioids (Genchi *et al.*, 2007) such that information generated can update or bridge knowledge gaps in this field. The One Health initiative is one platform where multidisciplinary approach could be taken to address this need. Therefore, a well-designed large scale survey targeting the definitive host, reservoirs and vectors using at least two diagnostic methods of high sensitivity and specificity including PCR and sequencing approach is worth consideration (Mar *et al.*, 2002; Bain *et al.*, 2008; Latrofa *et al.*, 2012b). To this end, zoologists, veterinarians, epidemiologists, physicians and environmentalists should come together to elucidate the epidemiology of canine filariosis in Nigeria.

CONCLUSION

The myriad of challenges in the Nigerian research arena notwithstanding, exercising restraint and caution through collaboration and seeking expert opinion before

publishing research findings is desirable to avoid misleading the research community. As it currently stands, the species of filarioid worms infecting dogs in Nigeria and their true prevalence are subject to debate. A comprehensive survey using at least two sensitive diagnostic approaches is needed to unravel the epidemiology of canine filarioids in Nigeria. The One Health platform is a good option to achieve this.

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