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Evaluation of Black-Headed Oriole *Oriolus brachyrhynchus* (Swainson, 1837) as Bioindicator of Arsenic Contamination Using Atomic Absorption Spectrometry (AAS)

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Authors' contributions

This work was carried out in collaboration between all authors. Authors FAE and POE designed the study author FAE performed the statistical analysis, wrote the first draft of the manuscript. Authors POE and BTT supervised the analysis of the study and paper work. All authors read and approved the final manuscript.

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ABSTRACT

An evaluation of Black-headed oriole Oriolus brachyrhynchus as bioindicator of arsenic contamination using Atomic Absorption Spectrometry (AAS) was carried out to determine the bioaccumulation level of arsenic concentration in the liver, skin, heart, adult feathers, nestling feathers, and eggs so as to ascertain which of these parts harbours more contaminants. A total of 30 birds were trapped from different farm locations in Buruku, Daudu and Adega using mist net and 150 of parts were obtained. A total of 9 eggs and 9 nestling were also collected from different nest from the study locations for investigation. We also examined the crops foraged on, and the water and soil of the environment. Summary statistics of Arsenic concentration (mg/kg) in selected parts were expressed using descriptive statistics and correlation. Arsenic concentration in feathers of the

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said species ranged between 1.29- 2.49 mg/kg with Daudu having the highest mean of 1.84 ± 0.42 mg/kg, whereas Buruku and Adega have below detection limit of 0.001 µg/L. Arsenic concentration in the liver of Black headed oriole ranged between 0.28 – 1.57 mg/kg with the birds in Daudu having the highest mean of 0.70 ± 0.47 mg/kg. Arsenic concentration in the skin ranged between 0.19 – 0.79 mg/kg with the birds in Daudu having the highest mean of 0.46 ± 0.21 mg/kg. Consequently arsenic concentration in carcass of Black headed oriole ranged between 0.28 – 3.42 mg/kg with the birds in Daudu having the highest mean of 2.47 ± 0.94 mg/kg. In addition, arsenic concentration in eggs and nestling feathers ranged between 0.04 – 0.05 mg/kg and 1.29 – 2.22 mg/kg with Daudu having highest arsenic concentration in both parts with mean of 0.05± 0.01 mg/kg and 1.63 ± 0.51 mg/kg respectively. Arsenic content of maize obtained from Daudu ranged between 0.46 - 0. 66 mg/kg with Daudu having the highest arsenic content with mean of 0.53 ± 0.11 mg/kg. In addition, arsenic content of soil ranged between 2.95 – 23.98 mg/kg with mean of 23.83 ± 0.14 mg/kg. Arsenic content of water ranged between 0.02 – 0.06 mg/kg. Therefore Daudu has the highest arsenic content in soil and water as a component of biological pathway for both crop plants and birds. The concentration of arsenic in different parts of the birds is a function of different time of exposure of individual bird trapped from different location, while arsenic levels in the feathers is an indicator of diet. It is imperative to study the potential mechanism by which arsenic cause diseases in wild birds which may be applicable to conservation of bird species to promote the conservation of wild birds and other animals especially species that are endanger and threaten. Therefore, analysis of mercury, lead, chromium and cadmium should be carried out to determine their accumulation in organs of Black headed oriole since these communities are agrarian communities.

Keywords: Arsenic; bioindicator; black-headed; evaluation; contamination.

1. INTRODUCTION

Arsenic is present in the environment due to its natural occurrence and human activities, for example agricultural and industrial activities. Arsenic pollution in any environment could be considered to be a potential threat to components of the ecosystem especially bird species that utilizes wetland. However, elevated contaminant levels may persist in sediments in some wetlands [1] were Black headed oriole forages.

With global change and increasing levels of industrial, commercial and agricultural contaminants it is important to study particular species living in a sensitive habitat [2] such as agricultural ecosystem for example Daudu, Buruku and Adega in Benue State, Nigeria. The types of fertilizer commonly used in these areas are Urea, Nitrogen-Phosphorous-Potassium (NPK), and Superphosphate [3]. Birds have been recognized as good indictors of environmental contamination because of their abundance, wide distribution, feeding at different trophic levels, and their long life span [4,5]. Arsenic occurs naturally in the soil in different types of rock and mineral ores such as copper (Cu), lead (Pb), cobalt (Co) and gold (Au) [6,7]. Mining activities, coal burning, pesticides and wood preservative application are the major anthropogenic sources of arsenic in the environment [6,8]. A number of studies have explored how regular pollutants destructively affect bird populations for instance, the increased use of pesticides [9] increases food production but with an increase in the level of arsenic contaminants in agricultural landscape.

Apart from that, it is also considered to be a serious hazard capable of affecting breeding performance and local survival of birds [10]. Arsenic is second to lead as one of the major heavy metal intoxication in birds which is an environmental toxicant and carcinogen. Liver is an important target organ of arsenic toxicity [11]. Arsenic is methylated by alternating reduction of pentavalent arsenic to trivalent and addition of methyl group from S-adenosylmethionine [11,12]. Dimethylarsinic acid (DMA) and Monomethylarsinic acid are the predominant metabolites of inorganic arsenic and are more toxic than the parent compound. It induces lipid peroxidation and Fe-dependent formation of reactive oxygen species (ROS) that leads to cellular damage and carcinogenesis [11,13] of various organs and tissues including skin, liver, lung and bladder. Arsenic acts to coagulate protein, forms complexes with coenzymes and inhibits the production of adenosine triphosphate during respiration [14] in birds.

Although the toxicology effects of arsenic are now more pronounced in recent time than ever [15], but basic evidence for the prospective side effects that changes community structure with promising pollutants, along with the propelling links between specific species and their functional-guild in terms of biotic responses remains limited in the study areas. The main objective of the study is to determine, weather the arsenic concentration in the various parts of the birds differs with location.

2. MATERIALS AND METHODS

2.1. Study Area

The study sites were located at Buruku on Latitude 7°27'35.6"N and Longitude 9°12'20.5"E in Buruku LGA, Daudu on Latitude 7° 55' 53.0" N and Longitude 8° 34' 53.9" E in Guma LGA and Adega on Latitude 7° 01'47.4" N and Longitude 8°15'28.0"E in Obi LGA of Benue State.

2.2 Sampling Technique

Using selective sampling technique which is a non- probability sampling techniques based on personal choice without statistical bias. The study sites are not a conservation area and the local people are predominantly peasant farmers. A total of 28 hectares of farmland area was selected in each of these study sites. The 28 ha sampling plot is further divided into four sampling units of 7 ha, where bird capturing activities took place and ethical permit was obtained from Department of Forest protection and Management Ministry of water resources and Environment in Makurdi, Benue State Nigeria. A mist net was set in each sampling unit and the net was checked after every 2-3 hours to search for a catch. Three replicates of nestling feathers and eggs collected from the tree species used as breeding ground by birds were stored in plastic sampling bottles.

Ten birds was trapped using mist net at the various locations. The birds were slaughtered and weighed before they were defeathered, the feathers were dried in an oven and kept in polyethene bags and sealed tightly. The defeathered birds are dissected to remove the skin, heart and liver which were kept in a polyethene bags and stored in a dessicator for arsenic analysis. Three replicates of rice, maize and guinea corn were also collected directly from different farmland as component of biological pathway for arsenic contamination in Black-headed oriole. Three replicates of water and soil samples were equally collected from the study areas for arsenic concentration investigation as component of biological pathway.

2.3 Sample Cleaning

The feather samples collected were cut into about 0.3 cm using a stainless steel scissors and first rinsed in ethanol, then washed three times in distilled water and then finally washed again in ethanol in accordance to the recommendation of International Atomic Energy Agency [16]. These was placed in crucibles and dried in an oven at 75°C ± 5°C for 25 minutes. About 0.2 mg of treated feather sample was weighed and stored in an inert plastic container of 10 cm^3 capacity, corked tightly and kept for arsenic analysis using a Flame Atomic Absorption Spectrometry.

The skin, heart, liver and the carcass after the vital parts are removed were dried in an oven equipped with circulation system at 60ºC for 48 h, and homogenized using a porcelain mortar. Approximately 0.2 mg of dry sample was treated with 7ml of concentrated nitric acid and heated for 20 min in the microwave oven, as described by [17]. The resultant solution was transferred into a 100 ml volumetric flask and made up to volume with distil water. The solution was stored at 4ºC in polyethylene bottles until arsenic was analyzed, using a Flame Atomic Absorption Spectrometry. The same procedure was followed for rice, maize, guinea corn, water and soil collected and digested for this study.

2.4 Procedure

Potassium Borohydride (KBH_4), carrier liquor and blank sample were connected into their respective sucking tubes. At the start of the hydride generator (connected to the main AAS), the solutions are automatically suck into the system where the hydride was produced and transmitted to the electric quartz absorption tube were it was detected and concentration recorded in mg/L. The whistling of the hydride generator signals the completion of the hydride production and hence the result was ready for reading. After proper calibration with the standard ranging from 1-10 mg/L, the blank was replaced with sample the concentration of arsenic was determined.

2.5 Quality Control

To ensure accuracy of results, two blanks were prepared for a avian parts, cereal and water namely; A and B. Blank A comprises of 7 ml of concentrated nitric acid and 2 ml of hydrogen peroxide which were heated for 30 minutes and the content is allowed to cool down to a room temperature then transferred into a volumetric flask and diluted to a final volume of 100 ml with distil water whereas, blank B was 100 ml of distil water only. The contaminants present in blank A and B were subtracted from the contaminants present in parts of the birds, crops and water collected and digested for this study to obtained the actual concentration of contaminants present.

Similarly, two blanks were also prepared for soil samples namely C and D. Blank C comprises of 7 ml of freshly prepared aqua-reqia (3 mL HNO_3 + 9 mL HCl) in the ratio 1:3 which was heated for 45 minutes and the content is allowed to cool down to a room temperature then transferred into a volumetric flask and diluted to a final volume of 100 ml with distil water whereas, blank D was 100 ml of distil water only. The contaminants present in blank C and D were subtracted from the contaminants present in all the soil samples collected and digested for this study to get the actual concentration of contaminants in soil.

In addition to further ensure accuracy, the AAS machine was calibrated using a known concentration of metals. These metals were run in the AAS machine to determine the absorbance and concentration. Calibration curves were obtained by plotting concentration against absorbance.

2.6 Experimental Design and Data Analysis

Using Randomized Complete Block Design (RCBD) the data was analyzed, where locations represented the blocks and the bird species the treatment. Pearson correlation was use to test the correlation between weight of parts and arsenic concentrations in parts ($P < 0.05$). The Summary statistics of Arsenic concentration (mg/kg) in selected parts of Black-headed orioles was express in mean values with their respective standard errors, minimum and maximum values and results were presented in tables and bar chart.

3. RESULTS

From our research work, a total of 168 parts were obtained from Black headed oriole trapped from different location in Benue state for arsenic concentration investigation. Table 1 shows the Summary statistics of Arsenic concentration (mg/kg) in selected parts of Black-headed orioles, expressed in mean values with their respective standard errors, minimum and maximum values. Arsenic concentration (mg/kg) in feathers of Black headed oriole ranged between 1.29 – 2.49 mg/kg with Daudu having the highest mean of 1.84 ± 0.42 mg/kg whereas Buruku and Adega having below detection limit of 0.001 µg/L using hydride generation-atomic absorption spectrophotometer (HG-AAS).

Arsenic concentration (mg/kg) in liver of Black headed oriole ranged between 0.28 – 1.57 mg/kg with Daudu having the highest mean of 0.70 ± 0.47 mg/kg whereas Buruku and Adega having below detection limit (Table 1).

Arsenic concentration (mg/kg) in the heart of Black headed oriole trapped from the 3 locations was below detection limit (Table 1). Apart from that, arsenic concentration for nestling feathers of birds trapped ranged between 1.29 – 2.22 mg/kg with Daudu having the highest mean of 1.63 ± 0.51, whereas Buruku and Adega was below detection limit (Table 1). Arsenic concentration (mg/kg) in skin of Black headed oriole ranged between 0.19 – 0.79 mg/kg with Daudu having the highest mean of 0.46 ± 0.21 mg/kg whereas, Buruku and Adega having below detection limit (Table 1).

Similarly, arsenic concentration for carcass of birds trapped ranged between 0.28 – 3.42 mg/kg with Daudu having the highest mean of 2.47 ± 0.94 whereas, Buruku and Adega was below detection limit (Fig. 1). In addition to that, arsenic concentration for eggs of bird species ranged between 0.04 – 0.05 mg/kg with Daudu having the highest mean of 0.05 ± 0.01, whereas Buruku and Obi was below detection limit (Table 1). There was a significant correlation (r^2 = 0.680) between arsenic concentration in the liver and body weight of carcass of Black headed oriole trapped from the study locations (Table 2).

A total of 27 crop samples were collected from different location (Table 3) for arsenic concentration investigation. Arsenic content (mg/kg) of maize foraged on frequently by Black headed oriole in Buruku ranged between 0.46 – 0.46 mg/kg with mean of 0.46 ± 0.00 mg/kg whereas, the arsenic content (mg/kg) of maize obtained from Daudu ranged between 0.46 – 0.66 with mean of 0.53 ± 0.11 mg/kg. However, Daudu has the highest mean of arsenic content whereas, Adega was below detection limit. In addition, the arsenic content (mg/kg) of rice in the three locations (Buruku, Daudu and Adega) was below detection limit (Table 3).

Similarly, the arsenic content (mg/kg) of guinea corn obtained from Buruku and Adega were also below detection limit whereas arsenic content for guinea corn collected from farmland in Daudu ranged between 0.20 – 0.21 with mean of 0.21 ± 0.01 mg/kg which was the highest arsenic content found in guinea corn from the study locations (Table 3).

Table 1. Summary statistics of Arsenic concentration (mg/kg) in selected parts of Black-headed orioles trapped from Benue State, Nigeria

Location and parts of Black-headed orioles	N	Mean ± SD	Minimum	Maximum
Buruku				
Feathers	10	**	-	-
Liver	10	**	-	-
Heart	10	**	-	-
Skin	10	**	-	-
Carcass	10	**	-	-
Eggs	3	**	-	-
Nestling feathers	3	**	-	-
Daudu				
Feathers	10	1.84 ± 0.42	1.29	2.49
Liver	10	0.70 ± 0.47	0.28	1.57
Heart	10	**	-	-
Skin	10	0.46 ± 0.21	0.19	0.79
Carcass	10	2.47 ±0. 94	0.28	3.42
Eggs	3	0.05 ± 0.01	0.04	0.05
Nestling feathers	3	1.63 ± 0.51	1.29	2.22
Adega				
Feathers	10	**	-	-
Liver	10	**	-	-
Heart	10	**	-	-
Skin	10	**	-	-
Carcass	10	**	-	-
Eggs	3	**	-	-
Nestling feathers	3	**	-	-

*** Not Detected*

Table 2. Correlation between arsenic concentrations in different parts and weight of different parts

	As CF (mg/kg)	**As CC (mg/kg)**	**As CL (mg/kg)**	**As CS (mg/kg)**	**WF (g)**	**WC (g)**	**WL (g)**	**WS (g)**
As CF (mg/kg)	1.00							
As CC (mg/kg)	0.471	1.00						
As CL (mg/kg)	0.547	0.680**	1.00					
As CS (mg/kg)	0.238	0.415	0.466	1.00				
WF (g)	-0.082	0.020	0.170	0.529	1.00			
WC (g)	-0.870	-0.486	-0.018	0.110	0.540	1.00		
WL (g)	-0.280	-0.337	-0.150	0.041	-0.272	-0.104	1.00	
WS (g)	-0.313	0.127	-0.220	-0.045	0.151	-0.348	-0.593	1.00

*** = (P < 0.05)*

Key:
WF =Weight of feather — As CF = Arsenic concentration in feather
WL = Weight of liver — As CC = Arsenic concentration in carcass
WC= Weight of carcass — As CL = Arsenic concentration in liver
WS = Weight of skin — As CS = Arsenic concentration in skin

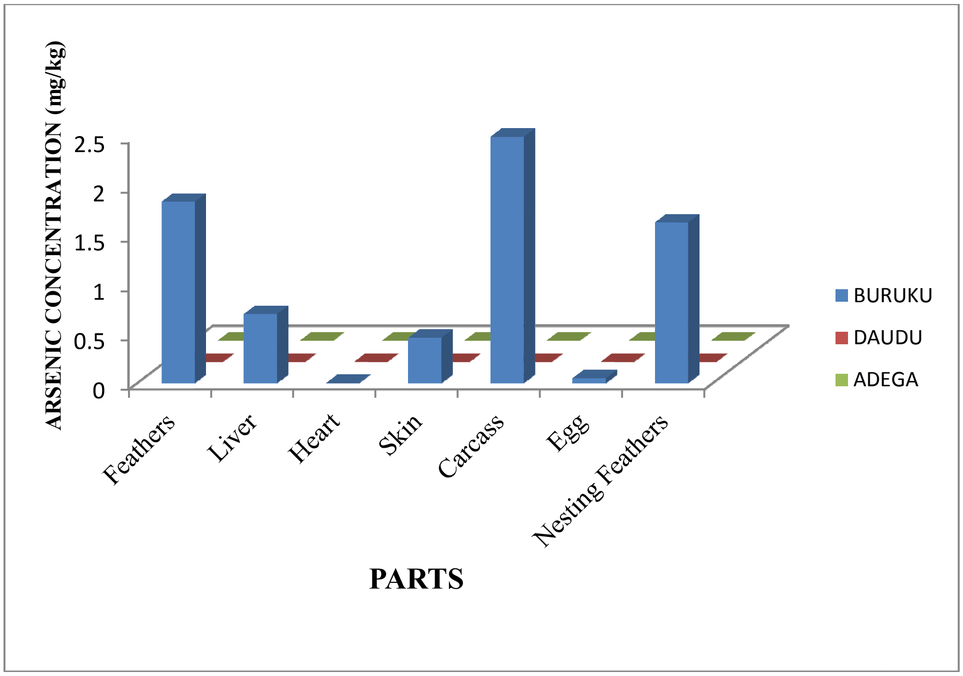

Fig. 1. Arsenic concentration in different parts

Table 3. Summary statistics of Arsenic concentration (mg/kg) in crops foraged on frequently by Black-headed orioles from the study locations

Locations and crops	N	Mean ± SD	Minimum	Maximum
Buruku				
Maize (*Zea mays*)	3	0.46 ± 0.00	0.46	0.46
Rice *Oryza sativa*	3	**	-	-
Guinea corn (*Sorghum bicolor)*	3	**	-	-
Daudu				
Maize (*Zea mays*)	3	0.53 ± 0.11	0.46	0.66
Rice *Oryza sativa*	3	**	-	-
Guinea corn (*Sorghum bicolor)*	3	2.08 ± 0.06	2.03	2.12
Adega				
Maize (*Zea mays*)	3	**	-	-
Rice *Oryza sativa*	3	**	-	-
Guinea corn (*Sorghum bicolor)*	3	**	-	-

*** Not detected*

Table 4. Summary statistics of Arsenic concentration in soil (mg/kg) and water (mg/L) analyzed from the study locations

Locations	N	Mean ± SD	Minimum	Maximum
Buruku				
Soil	3	3.11 ± 0.14	2.95	3.23
Water	3	0.03± 0.01	0.02	0.04
Daudu				
Soil	3	23.83 ± 0.14	23.71	23.98
Water	3	0.05 ± 0.01	0.04	0.06
Obi				
Soil	3	**	-	-
Water	3	**	-	-

*** Not detected*

A total of eighteen (18) samples were collected for both water and soil from different locations such as Buruku, Daudu and Adega in Benue State for arsenic concentration investigation. The arsenic content (mg/kg) of soil obtained from Buruku ranged between 2.95 – 3.23 mg/kg (Table 4), with mean of 3.11 ± 0.14 mg/kg whereas soil from Daudu ranged between 23.71 – 23.98 mg/kg with mean of 23.83 ± 0.14 mg/kg, but arsenic content of soil from Adega was below detection limit. Arsenic content mg/kg of water from Buruku ranged between 0.02 – 0.04 mg/L, with mean of 0.03 ± 0.01 mg/L and arsenic content mg/kg of water from Daudu ranged between 0.04 – 0.06 mg/L with mean of 0.05± 0.01 mg/L whereas water samples from Adega was below detection limit (Table 4). Therefore Daudu has the highest arsenic content in soil and water as a component of biological pathway for crop plants and birds.

4. DISCUSSION

The concentration of arsenic found in different organs is a function of different time of exposure of individual bird trapped from different location, while arsenic levels in the feathers is an indicator of diet during the period of feather growth and development which is not at variance with the findings of [18], because feathers are connected to blood vessels directly which makes it possible for arsenic to be incorporated into the keratin structure. Apart from that, it also represent circulating levels at the time of feather formation, they are indicative of internal levels at that time even in the nestling feathers and that of adult which is in agreement with findings of [2].

In this study we discovered that the concentration of arsenic in organs were not uniform this is due to difference in the rate of accumulation and the rate of elimination of the residues from the identified organs which is in agreement with the research work reported by [19]. Adult feathers and nestling feathers directly absorbed arsenic from the body (endogenous) and environment (exogenous). However, blood ladden with arsenic at the time of growing feathers and deposition of assimilated food in the liver are also responsible.

A higher accumulation of arsenic was found in the feathers, liver, nestling feathers and skin as a result of differences in bioaccumulation and biomagnifications of each organ. This is also liable for varying degrees of toxicity in the organs of Black headed orioles, because arsenic concentration in the said species has exceeded the permissible limit in animals including wild birds which ranges from 0.1 - 0.5 mg/kg in animal tissue [19]. The arsenic concentration of the heart was below detection limit from our research findings. However, the arsenic concentration in the liver was in consonance with work reported by [19] because the liver is considered to be storage organ and reserviour of toxic substances. Arsenic concentration in birds are known to be biomagnified and bioaccumulated over time and this is in agreement with the findings of [18,20,21]. The significant correlation between arsenic concentration in the liver and body weight of carcass of Black headed oriole trapped from the study location could be due to frequent consumption of cereal and water from the said area.

A number of studies indicate high levels of arsenic in water and cereal such as guinea corn and this could be a major contributor to inorganic arsenic in animal diets including avian species and this is in agreement with the research work reported by [22]. This study is not an exception because high level of arsenic was also recorded in Daudu which is considered to be potential source of contamination to birds of this locality because it has exceeded daily intake of 0.002 mg/kg, as reiterate by [23].

From this study the trace quantity of arsenic in the parts of birds was possibly due to environmental arsenic contamination of cereal crops such as maize and guinea corn utilized by the bird directly from the farm. Anthropogenic processes are also responsible for crop contamination because of continuous assimilation of small amounts of arsenic from the soil and water during the process of growth and development of crop plants.

Soil and water is considered to be the potential biological pathway for maize and guinea corn contamination. However, farmers in the area uses pesticides to improve crop productive, therefore natural rocks, pesticides application and annual flooding are the likely sources of arsenic contamination in this area since there are no big time industries.

In the present study, the differences found were large in magnitude compared to permissible limit. Black headed oriole are territorial, and they usually arrive on their breeding ground from March to June building nest before embarking on egg-laying but, their home range is very narrow during the breeding period which is more of agricultural field. Arsenic concentration in eggs could be acquired directly from feed consumed locally by the adult of the said species and it is delivered directly to the chick during the process of egg formation.

The basic effects of arsenic in birds have been reported such as teratogenic effects in birds. Arsenate could be teratogenic in the offspring of brooding birds. Equally too, arsenic has been shown to be capable of causing chromosome damage in bone marrow cells of birds as well [24]. The mechanism of arsenic genotoxicity is not clear, although several mechanisms have been proposed, including reactive oxygen species and the inhibition of deoxyribonucleic acid (DNA) repair [24,25]. In a study of the potential of arsenic compounds to act as promoters, a significant increase in the incidence of kidney tumours was observed in male birds [24].

Other studies using mice with specific genetic characteristics have shown carcinogenic effects [24,25], and these may be of value in studying the potential mechanism by which arsenic causes cancer which may be applicable to birds conservation, which may invariably promote the conservation of wild birds and other animals especially species that are endanger and threaten.

5. CONCLUSION AND RECOMMENDATION

It is imperative to study the potential mechanism by which arsenic cause diseases in wild birds which may be applicable to conservation of bird species to promote the conservation of wild birds and other animals especially species that are endanger and threaten. Therefore, analysis of mercury, lead, chromium and cadmium should be carried out to determine their accumulation in organs of Black headed oriole since these communities are agrarian communities.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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