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Prevalence, species distribution, diagnostic agreement and risk factors of intestinal parasitic infections among children attending a teaching hospital in Southeastern Nigeria

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Abstract
Intestinal parasitic infections remain a major cause of morbidity among children in developing countries. This cross-sectional study was to determine the prevalence, species distribution, diagnostic agreement between two microscopy techniques, and risk factors for intestinal parasitic infection among children aged 1–9 years attending Chukwuemeka Odumegwu Ojukwu University Teaching Hospital (COOUTH), Awka, Anambra State, Nigeria, over a six-month period. Stool specimens from 360 children were examined using saline wet mount and formol-ether concentration techniques, and a structured pretested questionnaire captured socio-demographic and environmental risk factors. From the 360 specimens, 134 (37.2%) were positive for at least one parasitic agent. *Ascaris lumbricoides* dominated (81.3% of positive findings), followed by *Trichuris trichiura* (9.7%), *Entamoeba histolytica* (8.2%), and *Giardia lamblia* (0.7%). The two diagnostic techniques showed substantial agreement (Cohen's Kappa = 0.799; observed agreement = 91.1%), but McNemar's test revealed a significant asymmetry ($p = 0.0001$), formol-ether concentration detected 27 cases missed by wet mount, compared with only 5 detected by wet mount alone, giving wet mount a sensitivity of 79.1% relative to formol-ether. On multivariate logistic regression, rural residence was independently associated with parasitic infection (adjusted OR = 1.96, 95% CI: 1.18–3.23, $p = 0.009$), while use of bottled drinking water was protective relative to borehole water (adjusted OR = 0.40, 95% CI: 0.19–0.82, $p = 0.012$). These findings demonstrate a substantial burden of intestinal parasitic infection among children attending COOUTH, dominated by *Ascaris lumbricoides*, and confirm that reliance on wet mount microscopy may lead to clinical under diagnosis. Incorporation of a concentration technique and saline wet mount alongside targeted water, sanitation and hygiene interventions in rural communities is recommended.

Keywords: Intestinal parasites, *Ascaris lumbricoides*, formol-ether, wet mount, risk factors

Introduction

Intestinal parasitic infections, comprising both protozoa and helminths, remain among the most prevalent infections of childhood in low- and middle-income countries and constitute a persistent public health problem in Nigeria (Aribodor *et al.*, 2012; Iwueze *et al.*, 2013) [5, 13]. Children between the ages of 1 and 9 years are particularly susceptible to intestinal parasitic colonisation because they are frequently exposed to contaminated environments through playing on the ground, poor handwashing habits, consumption of contaminated food and water, and close contact with infected individuals (Brooker *et al.*, 2015; Okeke and Eze, 2018) [6, 15].

Protozoan parasites such as *Entamoeba histolytica* and *Giardia lamblia*, and helminths such as *Ascaris lumbricoides*, *Trichuris trichiura* and hookworm, have all been reported among Nigerian children (Imakwu *et al.*, 2016; Iwueze *et al.*, 2013; Aribodor *et al.*, 2012 [5, 12, 13];). *Entamoeba histolytica*, the causative agent of amoebiasis, invades the intestinal lining and may cause abdominal pain, bloody stool and intestinal ulceration, while *Giardia lamblia* infection may result in malabsorption, abdominal discomfort, bloating and weight loss. Many infected children carry these organisms asymptomatically, sustaining silent transmission within communities (Imakwu *et al.*, 2016) [12]. Soil-transmitted helminths such as *Ascaris lumbricoides* and *Trichuris trichiura* are widely distributed in tropical with poor environmental sanitation. Eggs of these parasites are transmitted through contaminated soil, food and water (Ezeagwuna *et al.*,2009) [11]. Chronic infestation is associated with anaemia, malnutrition, poor appetite, growth retardation and impaired cognitive development in school-aged children (Ajjampur *et al.*, 2021; Mekonnen *et al.*, 2020; Pullan *et al.*, 2019) [3, 14, 16].

Accurate laboratory diagnosis remains effective clinical management and surveillance of intestinal parasites. Saline wet mount microscopy is the most widely used first-line technique because it is simple, rapid and preserves parasite motility, but its sensitivity for light-intensity infections is limited while concentration techniques such as formol-ether sedimentation are recommended to improve detection, particularly of protozoan cysts and lightly-shedding helminth ova (Cheesbrough, 2018; Endris *et al.*, 2013) [7, 10]. Many tertiary laboratories in Nigeria continue to rely on wet mount microscopy alone, this may lead to missed detection of parasitic organisms in low intensity specimens.

In Awka Metropolis of Anambra State, Nigeria, poor sanitation, inadequate access to clean water, and substandard hygiene practices remain prevalent, creating conditions favourable for parasite transmission (Afolabi *et al.*, 2023) [1]. However, hospital-based data on the species distribution, diagnostic performance and risk factors of intestinal parasitic infection among children in this locality are limited and are not current. This study therefore determined the prevalence and species distribution of intestinal parasitic infections, compared the diagnostic performance of saline wet mount against formol-ether concentration, and identified the socio-demographic and environmental determinants of parasitic positivity among children aged 1–9 years attending Chukwuemeka

Odumegwu Ojukwu University Teaching Hospital (COOUTH), Awka.

Materials and Methods

Study Area and Design

Cross-sectional descriptive study was conducted at Chukwuemeka Odumegwu Ojukwu University Teaching Hospital (COOUTH), a tertiary referral institution located in Amaku, Awka South Local Government Area of Anambra State, southeastern Nigeria. The hospital has a total bed capacity of about 440 and a functional paediatric department attending to a substantial number of children daily including those in Awka and environs. Data and specimen collection were carried out over a period of six months.

Ethical Considerations

Ethical approval was obtained from the Ethics Review Committee of COOUTH (Ref: COOUTH/HREC/Vol.11/FN:05/04848).

Informed Consent

Informed consent consents were obtained from the parents or guardians of each child prior to enrolment, and all data collected were kept confidential and used solely for research purposes and data collection. All data collected were kept confidential and used solely for research purposes.

Study Population, Sample Size and Sampling Technique

Eligible participants were children aged 1–9 years attending paediatric clinic of COOUTH, whose parents/guardians gave informed consent and who had not received antibiotic or antiparasitic treatment within the preceding two weeks. The minimum sample size was calculated using Cochran's formula for estimating proportions in a large population, based on an assumed prevalence of 38% in the south east (Egbewale *et al*., 2022) [9] (95% confidence level, margin of error = 0.05), A total of 360 360 children were consecutively recruited within the study period using a non-probability consecutive sampling technique.

Parasitological Examination of Stool Specimens

Stool specimens were examined using two techniques. Saline wet mount was performed by placing a drop of normal saline on a clean grease-free glass slide; mixing with a small portion of fresh stool, and examined under the x10 and x40 objectives of a compound microscope within 30 minutes of collection to preserve trophozoite motility (Zaman *et al.,* 2017) [17]. For formol-ether concentration technique, an estimated 2g of stool specimen was emulsified in 10% formalin, strained, mixed with diethyl ether, centrifuged at 2000rpm for 2minutes and the sediment examined microscopically using x10 and x40 magnification (Cheesbrough, 2018) [7].

Collection of data on Risk-Factors of intestinal parasitic infections

A structured, pre-tested, interviewer-administered questionnaire was used to collect socio-demographic and environmental data from parents/guardians, covering the child's age and sex, residence, parental education, source and treatment of drinking water, handwashing practice, presence of domestic animals, rotavirus vaccination history, recent history of diarrhoea, and household size.

Statistical Analysis

Data were entered into Microsoft Excel, cleaned, and analysed using IBM SPSS version 26.0. Descriptive statistics (frequencies and percentages) were used to analyse data on socio-demographic characteristics and parasite prevalence. Cohen's Kappa statistic and McNemar's test for paired discordance were used to assess agreement between the wet mount and formol-ether techniques. Chi-square tests of independence examined bivariate associations between categorical risk factors and parasitic positivity, and variables reaching significance were entered into a multivariable logistic regression model to identify independent determinants of parasitic infection. Statistical significance was compared at $p < 0.05$.

Results

The study population comprised 360 children, most aged 4–6 years (48.3%) and female (60.8%), giving a male-to-female ratio of approximately 1:1.6. Most children resided in urban areas (71.7%), while 28.3% were from rural communities (Table 1). The majority of parents/guardians had attained at least secondary education (57.2%), with the remainder having primary education only (42.8%).

Table 1: Socio-demographic characteristics of the children sampled

Characteristic	Category	N	%
Age group	1–3 years	70	19.4
	4–6 years	174	48.3
	7–9 years	116	32.2
Sex	Male	141	39.2
	Female	219	60.8
Residence	Urban	258	71.7
	Rural	102	28.3
Parent/guardian age	20–30 years	188	52.2
	31–40 years	156	43.3
	41+ years	16	4.4
Parent/guardian education	Primary	154	42.8
	Secondary	206	57.2
Number of children in household	1	34	9.4
	2	137	38.1
	3	87	24.2
	4	68	18.9
	5	34	9.4

Of the 360 stool specimens examined parasitic agents, *Ascaris lumbricoides* was similarly dominant, accounting for 109(81.3%), followed by *Trichuris trichiura* 13(9.7%), *Entamoeba histolytica* cysts 11(8.2%), and *Giardia lamblia* 1(0.7%) (Table 2).

Table 2: Species distribution of intestinal parasites identified in stool specimens (n = 134 positive specimens)

Positive species	Number of Parasites	Percentage (%)
Ascaris lumbricoides	109	81.3
Trichuris trichiura	13	9.7
Entamoeba histolytica	11	8.2
Giardia lamblia	1	0.7
Total	134	100.0

The two microscopy methods showed substantial overall agreement (Cohen's Kappa = 0.799; observed agreement = 91.1%). However, McNemar's test revealed that the disagreement between methods was significantly asymmetric (p = 0.0001). Formol-ether concentration detected 27 cases that wet mount missed, compared with only 5 cases detected by wet mount alone. This corresponds to a sensitivity of wet mount of only 79.1% and 97.8% relative to formol-ether concentration (Table 3).

Table 3: Diagnostic performance of saline wet mount relative to formol-ether concentration (n = 360)

Metric	Value
Observed agreement	91.1%
Cohen's Kappa	0.799 (substantial agreement)
Sensitivity of wet mount (vs. formol-ether)	79.1%
Specificity of wet mount (vs. formol-ether)	97.8%
McNemar's test statistic	5
McNemar's test p-value	0.0001

On bivariate analysis, two factors were significantly associated with parasitic positivity: residence (p = 0.005), with rural children showing a substantially higher positivity rate (49.0%) than urban children (32.6%); and number of children in the household (p = 0.038), with households having more than one child showing higher prevalence than single-child households (Table 4). No significant association was found for age group, sex, parental education, water source, water treatment, handwashing practice, presence of domestic animals, rotavirus vaccination status (p > 0.05).

Table 4: Association between residence and household size with parasitic positivity

Variable	Category	Number Sampled	Positive %	P-Value
Residence	Urban	258	84 (32.6)	0.0053
	Rural	102	50 (49.0)	
No. of children in household	1	34	6 (17.6)	0.0379
	2	137	55 (40.1)	
	3	87	40 (46.0)	
	4	68	21 (30.9)	
	5	34	12 (35.3)	

Statistically significant (p < 0.05)

On multivariable logistic regression adjusting for age, sex, residence, parental education, water source, water treatment, handwashing practice, rotavirus vaccination status, and household size simultaneously, residence remained significantly and independently associated with parasitic infection (adjusted OR = 1.96, 95% CI: 1.18–3.23, p = 0.009), indicating that children in rural areas had approximately twice the odds of parasitic infection compared with urban children, independent of other measured factors. Household size lost statistical significance after adjustment (adjusted OR = 0.98, p = 0.835). Use of bottled drinking water emerged as significant only in the adjusted model, and was associated with a lower risk of parasitic infection relative to borehole water (adjusted OR = 0.40, 95% CI: 0.19–0.82, p = 0.012) (Table 5).

Table 5: Multivariable logistic regression for independent determinants of parasitic positivity

Risk Factor	Category (vs reference)	Adjusted OR	95% CI	P-Value
Residence	Rural (vs. Urban)	1.96	1.18–3.23	0.009
Water source	Bottled water (vs. Borehole)	0.40	0.19–0.82	0.012
Water source	Tap water (vs. Borehole)	0.85	0.27–2.70	0.784
Sex	Male (vs. Female)	0.62	0.38–1.02	0.059
Age group	4–6 years (vs. 1–3 years)	0.83	0.46–1.50	0.537
Age group	7–9 years (vs. 1–3 years)	0.60	0.31–1.16	0.129
No. of children	per additional child	0.98	0.80–1.20	0.835

Significant determinant; OR: Odds Ratio; CI: Confidence Interval

Discussion

The overall parasitic positivity of 37.2% recorded in this study confirms that intestinal parasitic infection remains a substantial burden among children attending COOUTH, Awka. This is higher than the 23.6% intestinal parasite prevalence recorded among primary school children in a rural community in Kwara State, Nigeria (Amisu *et al.*, 2023) [4]. Likely reflecting differences between a hospital-based, partly symptomatic sample and a community-based asymptomatic school sample.

Ascaris lumbricoides dominated the parasitic isolates, accounting for 81.3% of positive findings. This is consistent with, though proportionally higher than, findings from Kwara State, Nigeria, where *A. lumbricoides* accounted for 50.8% of parasitic infections (Amisu *et al.*, 2023) [4]. Cross-sectional study of rural Ghanaian schoolchildren in which *A. lumbricoides* was identified in 57.4% of infected children (Dumevi *et al.*, 2026) [8]. The consistently high ranking of *A. lumbricoides* across these West African settings reflects the parasite's direct faecal–soil–oral transmission cycle, the durability of its ova in the environment, and its close association with open defecation and unsafe water use, all of which remain prevalent in both rural and peri-urban parts of Nigeria.

The comparatively low detection of *Giardia lamblia* in this study is notable when set against a recent nationwide, molecular (real-time PCR)-based survey of Nigerian children reporting substantially higher *Giardia* prevalence (Ajakaye *et al.*, 2026) [2]. This difference can best explained by diagnostic sensitivity rather than true biological variation: conventional wet mount and formol-ether microscopy, as used in this study, are known to under-detect protozoan cysts and trophozoites compared with molecular methods (Cheesbrough, 2018) [7]. The true burden of protozoan parasitism, such as giardiasis, in this population is therefore likely underestimated.

The two microscopy techniques showed substantial agreement (Cohen's Kappa = 0.799), but McNemar's test revealed a significant asymmetry in discordant pairs (p = 0.0001), with formol-ether concentration detecting 27 cases missed by wet mount, against only 5 detected by wet mount alone, giving wet mount a sensitivity of only 79.1% relative to formol-ether. The finding that wet mount alone would have missed a meaningful proportion of true positives is consistent with earlier work in Ethiopia, where single wet mount examination detected substantially fewer positive specimens than formol-ether concentration and Kato-Katz techniques applied to the same specimens (Endris *et al.*, 2013) [10]. These findings support standard parasitological guidance (Cheesbrough, 2018) [7] that a concentration technique such as formol-ether should not be omitted in favour of wet mount alone in routine diagnosis, due to the risk of missing light-intensity infections.

On multivariate analysis, rural residence remained independently associated with parasitic infection after adjustment for other factors (adjusted OR = 1.96, 95% CI: 1.18–3.23, $p = 0.009$), indicating that children in rural areas had approximately twice the odds of parasitic infection compared with their urban counterparts. This finding aligns closely with a Nigerian study in Kwara State in which rural dwellers was significantly associated with intestinal parasitic infection (Amisu *et al.*, 2023) [4]. Higher burden of intestinal parasitic infection recently documented among rural schoolchildren in Ghana, where inadequate sanitation and unsafe water access were identified as major drivers (Dumevi *et al.*, 2026) [8]. The protective association observed for bottled water use relative to borehole water (adjusted OR = 0.40, 95% CI: 0.19–0.82, $p = 0.012$) indicates that water source is an important transmission route for parasitic infection in this population, consistent with the wider literature on water, sanitation and hygiene (WASH) determinants of childhood parasitic disease in Nigeria (Afolabi *et al*., 2023) [1].

Conclusion

This study demonstrates that intestinal parasitic infections constitute a substantial and under-recognised burden among children attending COOUTH, Awka, with more than a third of children testing positive. *Ascaris lumbricoides* emerged as the predominant parasite, consistent with a faecal–soil–oral transmission cycle sustained by inadequate sanitation. The demonstrated superiority of formol-ether concentration over wet mount microscopy alone indicates that routine reliance on wet mount risks a clinically important rate of missed parasitic diagnoses. The independent association between rural residence, water source, and parasitic infection reaffirms that water, sanitation, and hygiene remain central, modifiable determinants of childhood parasitic infection in this environment. Public health interventions should specifically target rural households and communities relying on borehole water, including improved water treatment, safe water storage, and sanitation infrastructure. Routine deworming programmes and caregiver health education on hygiene and safe water practices should be sustained and expanded, particularly in rural communities.

Limitations

This study was conducted at a single tertiary hospital over a six-month period, which may not capture seasonal variation in parasite prevalence and limits generalisability beyond the COOUTH catchment population. Parasitological diagnosis relied on conventional microscopy rather than molecular or antigen-based methods, which are known to under-detect protozoan cysts and trophozoites. The low *Giardia lamblia* detection rate in this study, compared with PCR-based national surveys, suggests substantial underestimation of protozoan infection. The cross-sectional design allows only for the identification of associations and does not permit causal inference.

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