

Malaria, Typhoid, and PCV Variations: A Cross-Sectional Study in Akinima, Nigeria

Abstract

Malaria and typhoid fever remain major public health concerns in sub-Saharan Africa, particularly in Nigeria, where co-infections are common in resource-limited settings. This study investigated the prevalence and hematological impact of malaria, typhoid fever, and their co-infection among adults and children in Akinima, Ahoada West Local Government Area, Rivers State, Nigeria. A total of 191 participants were examined between August and November 2022: 44 (23.0%) adult males, 66 (34.6%) adult females, and 81 (42.4%) children. Malaria was diagnosed using Giemsa-stained blood films; typhoid fever was assessed using Widal agglutination tests; and packed cell volume (PCV) was determined via the microhematocrit method. Malaria prevalence was high in all groups: children (77.8%), adult males (77.3%), and adult females (74.2%). Typhoid fever was most prevalent among children (45.7%), followed by adult females (40.9%) and adult males (38.6%). Co-infection was highest in children (38.3%), indicating increased susceptibility in this age group. Hematological analysis showed that adult males had a normal mean packed cell volume PCV (45.0%), while adult females (34.0%) and children (34.2%) recorded lower mean values, suggesting anemia or hematologic compromise. These findings highlight significant differences in infection burden and hematological outcomes across age and sex. Children demonstrated the highest co-infection rates and lowest packed cell volume PCV values, underscoring their vulnerability. This study emphasizes the need for focused diagnostic, treatment, and preventive strategies, particularly for pediatric populations in rural, endemic communities.

Introduction

Malaria and typhoid fever are among the most prevalent infectious diseases in sub-Saharan Africa, often posing concurrent threats to public health, particularly in socioeconomically disadvantaged regions (17). Nigeria bears a significant share of this burden, accounting for approximately 27% of global malaria cases and a substantial proportion of typhoid-related morbidity and mortality (25). These diseases frequently coexist in endemic areas such as Akinima, located in the Ahoada West Local Government Area of Rivers State, where the tropical climate, poor sanitation, open drainage systems, and limited healthcare infrastructure contribute to their sustained transmission.

Malaria, predominantly caused by *Plasmodium falciparum*, remains a leading cause of morbidity and mortality in Nigeria. The parasite invades erythrocytes, causing hemolysis, anemia, and triggering a cascade of inflammatory responses (28). In contrast, typhoid fever, caused by *Salmonella enterica* serovar Typhi, a Gram-negative bacillus, spreads via the fecal-oral route, typically through the ingestion of contaminated water or food. Its pathogenesis involves intestinal invasion, systemic dissemination via the lymphatic and reticuloendothelial systems, and the release of endotoxins, which can impair immune function (5).

The clinical manifestations of malaria and typhoid fever often overlap, presenting with symptoms such as fever, headache, abdominal pain, and general malaise. This overlap can make differential diagnosis challenging in endemic regions (16). Misdiagnosis or delayed treatment is associated with an increased occurrence of severe complications, such as intestinal perforation in typhoid and cerebral malaria (9) or profound anemia in malaria, especially when both infections are present concurrently (9). This diagnostic difficulty is observed in resource-limited settings where access to confirmatory tools such as blood cultures, rapid diagnostic tests (RDTs), or PCR-based assays is limited. As a result, empirical treatment based on presumptive diagnosis is common, which can be a factor in antimicrobial resistance and therapeutic failures (20).

Children under five years of age are often observed to be vulnerable to these infections due to developing immune responses, hygiene practices, and exposure to unsanitary conditions (26). While adults may possess more robust immune systems, they can still experience the cumulative effects of recurrent or chronic infections, which may also be associated with hematological changes. Hematological parameters, such as hemoglobin concentration, packed cell volume (PCV), total and differential white blood cell counts, and platelet levels, are used as indicators of disease status and co-infection presence. Malaria is commonly associated with hemolytic anemia, thrombocytopenia, and leukopenia, while typhoid may be associated with neutropenia and bone marrow suppression (2) (17).

Observations of the hematological consequences of malaria-typhoid co-infection across different age groups are relevant for understanding disease patterns and developing public health strategies. Comparative studies that explore pediatric and adult responses to co-infection can indicate distinct observations, potentially guiding clinical decision-making and resource allocation, particularly in primary healthcare settings in communities like Akinima.

Therefore, this study aims to conduct a comprehensive comparative analysis of the hematological impact of malaria and typhoid co-infection among adults and children in Akinima, Ahoada West Local Government Area, Rivers State, Nigeria. By identifying observed differences in hematological parameters among these populations, the research presents information to understand the patterns of these co-endemic diseases.

Materials and Methods

A retrospective cross-sectional design was used, examining clinical data from 191 participants, comprising 44 adult males, 66 adult females, and 81 children aged 10 years and below. Laboratory investigations included malaria detection through microscopy, typhoid diagnosis via the Widal agglutination test, and PCV estimation using the microhematocrit method. Data were analyzed using descriptive statistics in SPSS and Excel.

Study Area and Population

This study was conducted between August and November 2022 at the Akinima Primary Health Center, the principal and most frequented healthcare facility in Ahoada West Local Government Area (LGA) of Rivers State, Nigeria. Akinima serves as the administrative headquarters of Ahoada West LGA and is situated in the northwestern part of Rivers State, within the Rivers West Senatorial District and the Port Harcourt metropolitan region (1). The town is observed to be impacted by recurrent annual flooding, which can be associated with public health challenges in the area (13).

Akinima experiences a tropical monsoon climate characterized by a hot and mostly cloudy dry season, and a warm, humid, and overcast wet season. The climate is generally oppressive year-round, with average temperatures ranging from 69°F to 88°F, rarely dropping below 62°F or exceeding 91°F (23). These climatic and environmental conditions can support the transmission of vector-borne and waterborne diseases such as malaria and typhoid fever.

Study Design

This study employed a descriptive cross-sectional design using a simple random sampling technique. Data collection was conducted over a four-month period, from August to November 2022, at the Akinima Primary Health Center in Ahoada West, Rivers State, Nigeria.

A total of 191 participants, comprising adult males, adult females, and children, were included in the study. All participants presented at the health center for medical evaluation and consented to laboratory testing for malaria parasitemia, typhoid antigen (Widal test), and packed cell volume (PCV). The demographic and hematological data collected served as the basis for the

comparative analysis of infection prevalence and hematological impact across the different population groups.

Ethical Consideration

Research authorization was obtained from the Ahoada West Local Government Authority and the relevant ethical review committee. Administrative approval was also secured from the management of Akinima Primary Health Center. Written informed consent was obtained from all adult participants prior to their inclusion in the study and before any blood sample was collected. For participants under the age of 18, informed consent was obtained from their parents or legal guardians. Participation in the study was entirely voluntary, and all respondents were assured of the confidentiality and anonymity of their responses in accordance with ethical standards for human subject research.

Biodata of the Participants and Blood Collection

The study population comprised 44 adult males, 66 adult females, and 81 children who presented at the Akinima Primary Health Center for medical consultation and were clinically observed for suspected malaria, typhoid fever, or anemia between August and November 2022. Recruitment was conducted through voluntary participation. Informed consent was obtained from all adult participants prior to their inclusion in the study. For minors, written consent was obtained from their parents or legal guardians. Participation was entirely voluntary, and participants retained the right to decline or withdraw at any point without prejudice.

Sociodemographic information, including age, sex, place of residence, educational level, occupation, and marital status, was collected, along with clinical symptoms such as fever, headache, joint pain, dizziness, fatigue, chills, vomiting, and diarrhea. Data on medical history and treatment practices, including the use of antimalarial drugs, antibiotics, or traditional/herbal remedies, were also recorded. These data were gathered using a pretested structured questionnaire administered through face-to-face interviews by trained personnel. Following the interview, 2mL of venous blood was aseptically collected from each participant by qualified phlebotomists using sterilized EDTA-anticoagulated tubes Falcon brand. Strict aseptic procedures were maintained throughout the sample collection process to ensure both sample integrity and participant safety. All samples for packed cell volume (PCV) analysis were processed within one hour of collection.

Sample Analysis

Malaria Parasite Detection Using Thick and Thin Blood Films

The diagnosis of malaria was carried out through microscopic examination of Giemsa-stained thick and thin blood films using an oil immersion objective lens ($\times 100$). Both film types were stained with 10% rapid Giemsa stain (SD Biotec, Inc., USA) for 15 minutes. The thick films were used for parasite detection, while thin films facilitated Plasmodium species identification. Parasitemia was quantified by counting the number of parasites per 200 white blood cells on the thick film, assuming an average leukocyte count of 8,000/ μL of blood (27). A minimum of 100 high-power fields was examined before a slide was reported as negative, in accordance with established protocols (Edit et al., 2016). If the thick smear tested positive for malaria parasites, species identification was performed on the corresponding thin smear based on morphological characteristics, using standard identification tables as described (8).

Widal Test

Typhoid fever was diagnosed using both semi-quantitative (rapid slide agglutination) and quantitative (tube agglutination) methods, following the procedure described (22). Initial screening was performed using the rapid slide agglutination test with commercially available Salmonella typhi antigen kits (Salmonella O, and H antisera). The test utilized somatic (O) and flagellar (H)

antigen reagents, specifically, TO and TH for detecting antibodies against *S. typhi*. Additional antisera (AO, BO, CO, AH, BH, and CH) were employed for identifying *S. paratyphi* A, B, and C. An antibody titer of $\geq 1:80$ against the TO and TH antigens of *S. typhi* was considered indicative of a positive result, while titers below this threshold ($< 1:80$) were interpreted as negative. All samples that tested positive during the slide test were subjected to confirmatory testing using the standard tube agglutination method. Each positive sample was tested in duplicate to ensure accuracy, consistency, and reproducibility of the results.

Packed Cell Volume (PCV) Determination

The packed cell volume (PCV) of each participant was measured using the microhematocrit method, following the standard procedure outlined (7). Approximately 2–3 drops of thoroughly mixed EDTA-anticoagulated venous blood were drawn into plain, heparin-free capillary tubes up to two-thirds of their length. One end of each capillary tube was sealed with plasticine, and the tubes were then placed in a microhematocrit centrifuge and spun at 11,000 revolutions per minute (rpm) for 5 minutes.

Following centrifugation, the PCV was determined using a microhematocrit reader by aligning the interfaces of the plasma, buffy coat, and red blood cell layers. The PCV values were interpreted based on standard hematological reference ranges:

Adult males: 40%–54%

Adult females: 35%–45%

Children under 10 years: 34%–40%

Statistical Analysis

Data were analyzed using Microsoft Excel and SPSS (Statistical Package for the Social Sciences) version 22. Descriptive statistics, including frequencies, percentages, means, and standard deviations, were used to summarize demographic characteristics and laboratory results.

Inferential statistical tools employed included the Chi-square test for assessing associations between categorical variables, independent sample t-tests for comparing means between two groups, and one-way ANOVA for comparisons across more than two groups. A 95% confidence interval (CI) was adopted, and statistical significance was set at $p < 0.05$.

Results

This study assessed the prevalence of malaria parasitemia (MP), typhoid antigen presence, and packed cell volume (PCV) among three distinct demographic groups: adult males, adult females, and children aged 10 years and below. The analysis focused on these three key parameters to evaluate infection patterns and their associated hematologic characteristics across these groups.

Table 1. A total of 191 participants were included in the study, comprising three distinct demographic groups: 44 adult males, 66 adult females, and 81 children aged 10 years and below. The adult male group included participants aged between 30 and 50 years, with a mean age of 40 ± 10 years, and consisted entirely of males. The adult female group included individuals aged 20 to 40 years, with a mean age of 30 ± 10 years, and consisted entirely of females. The children's group included participants aged 1 to 10 years, with a mean age of 5 ± 5 years. This group was composed of 31 males and 50 females, reflecting a higher number of female children. Overall, the study population had 75 males and 116 females, providing a basis for assessing observed

characteristics related to gender, age group, and the hematological aspects of malaria, typhoid, and their co-infection.

Demographic Group	Sample Size (n)	Age Range (Years)	Mean Age \pm SD (Years)
Adult Males	44	30–50	40 \pm 10
Adult Females	66	20–40	30 \pm 10
Children (\leq 10 years)	81	1–10	5 \pm 5
Total	191	—	—

Table 2. presents the prevalence of malaria among three different demographic groups: adult males, adult females, and children in Akinima, Ahoada West, Rivers State, Nigeria. Adult Males (n = 44): Out of the 44 adult males, 34 individuals (77.3%) tested positive for malaria, while 10 individuals (22.7%) tested negative. Adult Females (n = 66): Among the 66 adult females, 49 individuals (74.2%) tested positive for malaria, and 17 individuals (25.8%) tested negative. Children (n = 81): In the group of 81 children, 63 individuals (77.8%) tested positive for malaria, while 18 individuals (22.2%) tested negative. The highest prevalence of malaria was observed among children. The table indicates that malaria is present across all demographic groups, with children showing the highest rate of observed infection. The differences in observed prevalence between adult males, adult females, and children are small. These results describe the distribution of malaria in the study population.

Group	Sample Size (n)	Malaria Positive (n, %)	Malaria Negative (n, %)
Adult Males	44	34 (77.3%)	10 (22.7%)
Adult Females	66	49 (74.2%)	17 (25.8%)
Children	81	63 (77.8%)	18 (22.2%)

Table 3. presents the prevalence of typhoid antigen among the three demographic groups: adult males, adult females, and children in Akinima, Ahoada West, Rivers State, Nigeria. Adult Males (n = 44): Among the 44 adult male participants, 17 (38.6%) tested positive for typhoid antigen, while 27 (61.4%) tested negative. Adult Females (n = 66): Of the 66 adult female participants, 27 (40.9%) were typhoid-positive, and 39 (59.1%) were typhoid-negative. Children (n = 81): Out of the 81 children included in the study, 37 (45.7%) tested positive for typhoid antigen, while 44 (54.3%) tested negative. This group showed the highest prevalence of typhoid antigen. Overall, the data indicate that typhoid antigen is present across all demographic groups, with children showing the highest observed prevalence.

Table 3. Typhoid Antigen Prevalence Across Demographic Groups

Group	Sample Size (n)	Typhi Positive (n, %)	Typhi Negative (n, %)
Adult Males	44	17 (38.6%)	27 (61.4%)
Adult Females	66	27 (40.9%)	39 (59.1%)
Children	81	37 (45.7%)	44 (54.3%)

Table 4 shows the Analysis and Interpretation: Adult males recorded a mean packed cell volume (PCV) of 45.0%, which falls within the normal hematological range for their group. In contrast, adult females had a mean PCV of 34.0%, which is below their normal reference range. This observation is consistent with the abstract, indicating lower PCV values among adult females. Children exhibited a mean PCV of 34.2%, which is at the lower threshold of their normal pediatric range (34%–40%). This observation is consistent with the abstract's statement regarding lower PCV values in children. Overall, adult males showed the highest mean PCV (45.0%), while adult females and children had lower mean values (34.0% and 34.2%, respectively). These observed differences present the varying PCV distributions among females and children in the study population.

Table 4. Packed Cell Volume Analysis by Demographic Group

Group	Mean PCV (%)	Normal Range	Observation
Adult Males	45.0%	40–54%	Within normal range
Adult Females	34.0%	35–45%	Below normal reference range
Children	34.2%	34–40% (1–10 years)	At the lower threshold of normal range

Table 5 is based on the dataset of 44 adult males, 66 adult females, and 81 children, the analysis of malaria-typhoid co-infection revealed distinct patterns across the demographic categories. These observations are consistent with reports (10). Interpretation: Children exhibited the highest rate of malaria-typhoid co-infection, with a prevalence of 38.3%. Adult females recorded the second highest prevalence at 34.8%. Adult males showed the lowest co-infection rate at 27.3%. Overall, the results show that malaria and typhoid co-infection occur most frequently among children.

Table 5. Prevalence of Malaria-Typhoid Co-infection by Demographic Group

Group	Sample Size (n)	Co-infected (n)	Co-infection Prevalence (%)
Adult Males	44	12	27.3%
Adult Females	66	23	34.8%
Children	81	31	38.3%

Discussion

The study's findings on malaria parasitemia, typhoid antigen prevalence, and packed cell volume (PCV) values provide insights into the observed disease burden across different age and sex groups in Akinima, Ahoada West, Rivers State, Nigeria. The results highlight the public health characteristics presented by these infectious diseases.

Malaria Parasitemia

The data revealed a high prevalence of malaria across all demographic groups, with the highest rate observed among children (77.8%), followed by adult males (77.3%) and adult females (74.2%). The present study is in keeping with the findings (18), which revealed that malaria was prevalent across all age groups. Among these groups, children had the highest prevalence of malaria, followed by adults. Also in agreement with the present study are the findings (17), which indicate that malaria was prevalent across all groups but most common in adult females, followed by children. This finding is consistent with previous research indicating that children may experience more severe outcomes such as anemia (24) (14). The observed highest prevalence in children supports the general notion that this age group is more vulnerable to the disease, as partial immunity to malaria develops gradually with age and repeated exposure to the parasite.

Aside from the citations from previous studies, the high prevalence of malaria across all demographic groups in the present study can also be attributed to the endemic nature of the disease in the region. Breeding sites of the mosquito vector, such as the Akinima annual flooding, river overflowing, water retained in plant axils, and stagnant water in drainage systems, are common in the area. According to the (6), after feeding on human or animal blood, the female *Anopheles* mosquito rests for a few days to digest the blood and then lays eggs in water, where the larvae develop into adult mosquitoes. Furthermore, the female mosquito tends to bite at dusk or during the night due to its nocturnal nature, as light suppresses its activity. The local environment in Akinima is conducive to nighttime mosquito activity, possibly due to poor or inconsistent lighting. This lack of adequate light sources may have contributed to the widespread exposure and high malaria prevalence observed across the different demographic groups.

Typhoid Antigen Prevalence

The study also demonstrated a notable presence of typhoid antigen across all groups, with the highest prevalence in children (45.7%), followed by adult females (40.9%) and adult males (38.6%). This trend aligns with the study (12) and describes that children are observed to be vulnerable to typhoid fever due to factors such as hygiene practices, exposure to contaminated water and food, and developing immunity (12)(24). On the other hand, the study is not in keeping with the finding of (17) wherein the prevalence was lower across all groups and gender prevalence was equal. This can be explained on the basis that the previous study relied on hospital records as compared to the present study. Furthermore, the findings of the present study is in contrast with the report (18), which showed that young adults irrespective of their gender, had the highest prevalence rate of typhoid fever. This is suggestive of individuals within these groups patronizing restaurants where they buy food from food vendors which may have not been prepared in a hygienic environment.

Packed Cell Volume (PCV) and Anemia

Analysis of PCV values showed that PCV was lowest among adult females (mean PCV 34.0%) and children (mean PCV 34.2%), compared to adult males (mean PCV 45.0%). These results correspond with the observed prevalence of malaria and typhoid infections. The destruction of red blood cells by malaria parasites and the hematologic observations associated with chronic typhoid infections (such as bone marrow suppression) are factors that can contribute to changes in PCV (4) (3) & Brundha, 2020; (15). In adult females, additional factors like menstrual blood loss and dietary iron intake may also be associated with PCV levels. Children are

observed to have additional considerations due to nutritional factors, physiological demands during growth, and a higher burden of infections.

Co-infection Prevalence and Hematological Impact

The prevalence of malaria-typhoid co-infection was highest in children (38.3%), followed by adult females (34.8%) and adult males (27.3%). These findings are consistent with those (18), who reported the highest co-infection rate (37.5%) in children. The present study also agrees with (19), who recorded the highest co-infection rate (11%) among children. In contrast, these results differ from those of (17), who found that females exhibited a higher rate of co-infection compared to both males and children. Similarly, (16) also reported a higher co-infection rate (30.3%) among females. The higher prevalence in females, as reported by these studies, may be attributed to their increased exposure due to spending more time outdoors and having limited access to clean drinking water.

Meanwhile, the present study attributes the higher co-infection rate in children to their more pronounced clinical symptoms and lower PCV levels, which reflect greater vulnerability. This study presents a comparative analysis of the prevalence and hematological effects, specifically on packed cell volume (PCV) of malaria, typhoid fever, and their co-infection among adults and children in Akinima, Ahoada West, Rivers State, Nigeria. Recognizing the endemic nature of both diseases in sub-Saharan Africa, the research aimed to identify infection patterns across age and gender and to evaluate how co-infection correlates with PCV levels.

The results showed high malaria prevalence across all groups: adult males (77.3%), adult females (74.2%), and children (77.8%). Typhoid fever was also notable, with children again showing the highest positivity rate (45.7%), followed by adult females (40.9%) and adult males (38.6%). Co-infection was most frequent in children (38.3%), followed by adult females (34.8%) and adult males (27.3%). Hematological analysis revealed differences in PCV values. Adult males had the highest mean PCV (45.0%), while adult females (34.0%) and children (34.2%) exhibited lower mean PCV values. This trend indicates lower PCV levels among children and adult females, and co-infection was observed to be associated with reduced PCV values. The findings illustrate the co-occurrence of malaria and typhoid and their observed relationship with PCV, particularly among children.

Aside from the conclusions already stated, some of which are supported by previous studies, anemia is a common finding in both malaria and typhoid fever due to overlapping mechanisms. Both diseases suppress bone marrow activity, resulting in decreased red blood cell production and, consequently, anemia. However, typhoid fever has an additional mechanism that may explain the development of anemia: hemophagocytosis, a condition where activated macrophages engulf red blood cells, white blood cells, and platelets. This process further contributes to the observed hematological decline, especially in co-infected individuals.

In addition to positivity rates, malaria parasitemia was graded semi-quantitatively using a conventional microscopy-based scoring system (+, ++, +++, +++++), a widely accepted method for estimating parasite density in field settings (7)(21)(11). This grading, as shown in Table 5, serves as a proxy for parasite density and provides insight into infection intensity. A notable observation is the trend of lower PCV values in participants with higher parasitemia scores, particularly among children and adult females. Although this study did not statistically correlate parasite density scores with PCV due to its descriptive design, future analyses may explore this relationship to better understand the hematologic burden of varying infection intensities.

Conclusion

This study provides a comprehensive analysis of malaria parasitemia, typhoid antigen prevalence, and packed cell volume (PCV) levels across three demographic groups, adult males, adult females, and children in Akinima, Ahoada West, Rivers State, Nigeria. The findings reveal

that malaria is highly prevalent in all groups, with children exhibiting the highest infection rate (77.8%), followed by adult males (77.3%) and adult females (74.2%). This indicates that while malaria is widespread, children are observed to have the highest infection rates. Similarly, typhoid antigen prevalence was most common among children (45.7%), showing their higher observed prevalence. Adult females had a slightly higher typhoid rate (40.9%) than adult males (38.6%). Packed cell volume (PCV) analysis showed that adult males maintained PCV values within the normal range (mean PCV = 45.0%), whereas adult females (34.0%) and children (34.2%) displayed lower mean PCV levels, suggesting lower PCV observations in these groups.

Furthermore, co-infection analysis demonstrated that children had the highest malaria-typhoid co-infection rate (38.3%), followed by adult females (34.8%) and adult males (27.3%). This pattern presents the observed vulnerability of children, who show not only higher infection rates but also more pronounced hematologic observations when co-infected. This study presents the dual burden of malaria and typhoid fever in the population, particularly among children and adult females, and describes the observed associations.

Limitations

This study is the use of an assumed average white blood cell (WBC) count of 8,000 cells/ μ L for estimating malaria parasite density. While this approach aligns with standard protocols in resource-limited settings, it may not reflect individual variations in leukocyte counts among participants. The unavailability of equipment and reagents for conducting actual WBC counts at the study site restricted our ability to obtain more precise parasitemia measurements. This may have introduced a degree of variability in parasite quantification. Future studies should aim to include individualized hematological parameters to enhance diagnostic accuracy and data reliability.

Table 6. Raw Data from the Cross-Sectional Study Showing Malaria Parasite Load, Typhoid Antigen Titers, and Packed Cell Volume (PCV) Variations Among Adult Males, Adult Females, and Children in Akinima, Nigeria.

Malaria parasite presence is reported as Not Seen, +, ++, +++, or +++++. Typhoid infection is indicated by Widal agglutination titers (1:20 to 1:360). Packed Cell Volume (PCV) is expressed in percentage (%), with normal reference ranges as follows: Adult Males (40–54%), Adult Females (35–45%), and Children aged 1–10 years (34–40%)

TEST	ADULT MALE			ADULT FEMALE			CHILDREN 1-10yrs		
	MP	Typhi	PCV	MP	Typhi	PCV	MP	Typhi	PCV
NR			40-54%			35-45%			34-40%
1	+	20	41%	+	20	36%	++	160	35%
2	Not seen	20	45%	Not seen	20	35%	+	20	37%
3	+	20	45%	++	80	30%	++	80	30%
4	+	20	40%	++	20	34%	++	20	34%
5	+	20	45%	++	20	34%	++	160	33%
6	+	20	46%	+	20	35%	+	20	39%

7	+	20	47%	++	20	31%	++	20	32%
8	+	20	45%	++	80	33%	++	160	33%
9	Not seen	20	50%	+	20	38%	Not seen	20	36%
10	++	160	55%	++	20	35%	++	20	35%
11	++	80	48%	++	20	35%	++	20	36%
12	Not seen	20	49%	Not seen	80	38%	+	160	33%
13	++	80	41%	++	20	29%	++	20	29%
14	++	80	47%	++	20	30%	++	20	34%
15	++	80	43%	++	20	31%	++	20	30%
16	++	80	41%	++	20	30%	++	20	30%
17	++	80	46%	++	20	33%	++	20	31%
18	++	80	46%	++	20	34%	++	20	34%
19	++	80	40%	++	20	34%	++	20	34%
20	+++	360	39%	+++	120	28%	++	360	33%
21	++	80	45%	++	20	37%	++	20	36%
22	Not seen	80	40%	Not seen	80	35%	Not seen	80	35%
23	Not seen	80	48%	+	80	35%	Not seen	80	35%
24	Not seen	80	40%	Not seen	80	35%	++	160	33%
25	Not seen	20	51%	+	80	35%	Not seen	80	40%
26	+	20	47%	Not seen	20	35%	Not seen	20	35%
27	+	20	46%	+	20	36%	+	20	36%
28	+	20	35%	+	20	38%	+	20	38%
29	+	20	46%	+	20	42%	++	20	36%
30	+	20	45%	+	20	38%	+	20	38%
31	+	20	38%	+	20	36%	Not seen	20	36%
32	+	20	40%	Not seen	20	36%	++	20	32%
33	+	20	47%	+	80	35%	+	80	35%
34	+	20	49%	+	160	33%	+	80	33%
35	+	20	48%	+	80	56%	+	160	33%
36	+	20	45%	+	20	58%	+	20	30%
37	+	20	49%	+	80	35%	+	160	33%
38	+	20	48%	Not seen	80	30%	Not seen	80	30%
39	Not seen	20	45%	+	20	35%	++	20	35%
40	Not seen	20	47%	++	80	34%	++	160	32%
41	Not seen	20	42%	++	80	30%	++	80	30%
42				++	20	39%	++	160	31%
43				+	80	35%	Not seen	80	35%

UNDER PEER REVIEW

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

CONSENT

As per international standards or university standards, Participants' written consent has been collected and preserved by the author(s).

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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