Open Forum Infectious Diseases
MAJOR ARTICLE

Re-emergence of Diphtheria in Guinea: An Outbreak Analysis of Vaccination and Disease Control Perspectives

Alpha Kabiné Keita,[1,2] Abdoul Karim Soumah,[1] Thibaut Armel Chérif Gnimadi,[1] Abass Kande,[1,2] Kadio Jean Jacques Olivier Kadio,[1] Haby Diallo,[1] Mariama Cisse,[1] Joel Ballè Koivogui,[1] Djiba Kaba,[1,2] Salifou Talassone Bangoura,[1,2] Abdoulaye Toure,[1,2] Florence Fenollar,[3,4] Oleg Mediannikov,[3,5,6] and Alpha Kabinet Keita[1,2,7]

[1]Guinea Infectious Diseases Research and Training Center, Gamal Abdel Nasser University of Conakry, Conakry, Republic of Guinea, [2]Faculty of Health Sciences and Techniques, Gamal Abdel Nasser University, Conakry, Republic of Guinea, [3]Aix-Marseille University, IHU-Méditerranée Infection, Marseille, France, [4]Aix-Marseille Univ, SSA, AP-HM, RITMES, Marseille, France, [5]Aix-Marseille Univ, AP-HM, MEPHI, Marseille, France, [6]Developpement Research Institute/IRD, French Ministry of Europe and Foreign Affairs, Marseille, France, and [7]TransVIHMI, University of Montpellier, National Institute of Healthand Medical Research/INSERM, Development Research Institute/IRD, Montpellier, France

Background. This study presented the key characteristics of patients who tested positive for diphtheria during the outbreak in the Republic of Guinea in 2023 and assessed the influence of some risk factors on disease development.

Methods. The clinical diagnosis of diphtheria was confirmed by detecting diphtheria toxin genes in nasopharyngeal samples collected from suspected patients via 2 reverse transcription-quantitative polymerase chain reaction tests. Bivariate analyses with the χ^2 test and the Fisher's exact test were conducted to explore possible associations between diphtheria positivity and various sociodemographic, clinical, and exposure factors.

Results. In total, 444 samples obtained from suspected cases were analyzed. In 90 (20.3%) cases, the condition was confirmed using quantitative polymerase chain reaction, with an overall fatality rate of 8.9% ($n = 8$). On average, deaths occurred 2 days after admission, with 6 (75.0%) of 8 (6 girls and 2 boys) deaths recorded within the first 3 months after the epidemic onset. The clinical characteristics included sore throat (91%), fever (90%), whitish throat membrane (83%), throat redness (81%), and dyspnea (28%). The risk factors were age <15 years, no prior vaccination, and contact with a patient with diphtheria. A whitish throat membrane and dyspnea were significantly associated with diphtheria positivity.

Conclusions. This study emphasized that diphtheria remains a major and potentially fatal disease, despite vaccination and early symptom recognition. The identification of characteristic signs—particularly a whitish throat membrane and dyspnea—is important for reducing disease severity and mortality.

Keywords. epidemic surveillance in Guinea; re-emergence of diphtheria; vaccine-preventable disease.

Diphtheria is a potentially fatal infectious disease. However, it can be prevented with vaccination. The term, which originated from the Greek word "diphtheriae," refers to the pseudomembrane that forms at the infection sites [1, 2]. The disease is caused by *Corynebacterium diphtheriae*, a Gram-positive actinobacterium with a high G+C content. Its incubation period is 2–5 days, and the condition is initially characterized by sore throat and fever. Severe cases involve toxin-mediated production of a characteristic gray or whitish pseudomembrane in the throat [3–5].

Clinically, diphtheria presents in pharyngeal and cutaneous forms, commonly caused by the toxigenic strains of *C. diphtheriae* and rarely by *Corynebacterium ulcerans* and *Corynebacterium pseudotuberculosis*, collectively leading to thousands of deaths annually worldwide [4, 6, 7].

Despite achieving a greater control over the disease globally, several countries in Africa, Europe, Asia, and America continue to experience some level of re-emergence and increase in diphtheria cases [8]. According to World Health Organization data, 84% of children worldwide were fully vaccinated against diphtheria during early childhood. Meanwhile, the remaining 16% were either unvaccinated or not fully vaccinated, with a high risk of contracting the disease and a case fatality rate of 5%–10% [9]. Generally, additional clinical and biological factors contribute to the development of diphtheria. A better understanding of these risk factors would facilitate a more precise identification of vulnerable populations and support the development of more effective interventions, thereby reducing disease burden [10].

In 2023, a diphtheria outbreak emerged in north-eastern Guinea, where the vaccination coverage among children aged 0–23 months was only 54% [11]. By November 2024, the National Agency for Food Safety reported 6331 suspected cases

Received 12 March 2025; editorial decision 18 August 2025; accepted 27 August 2025; published online 30 August 2025

Correspondence: Alpha Kabinet Keita, PhD, Gamal Abdel Nasser University of Conakry, Guinea Infection Research and Training Center, BP: 6629P Dixinn, Conakry, Republic of Guinea (alpha-kabinet.keita@cerfig.org).

https://doi.org/10.1093/ofid/ofaf527

of diphtheria and 162 deaths since the first case was biologically confirmed on 20 July 2023 [11]. Importantly, the 2018 Demographic and Health Survey showed that only 24% of children aged 12–23 months had been fully vaccinated in Guinea. Meanwhile, 22% of children from the same age group did not receive vaccination at all [12]. This finding emphasizes the issue of under-immunization against vaccine-preventable diseases such as diphtheria within the population.

Due to this alarming situation and with the aim to develop an appropriate epidemiological response, the current study assessed the main characteristics of confirmed diphtheria cases and the influence of some risk factors on disease development.

MATERIALS AND METHODS

Data Collection and Study Design

This study conducted a secondary analysis of data gathered by health workers during the admission of patients with suspected diphtheria at epidemic treatment centers in the health districts that reported cases from July 18, 2023, to June 18, 2024. These data, which included sociodemographic, clinical, and epidemiological information on suspected cases, were obtained directly from notification forms. Nasopharyngeal or oropharyngeal swab samples were also collected for the laboratory confirmation of the diagnosis using 2 real-time quantitative polymerase chain reaction (RT-qPCR) tests, which can detect bacterial DNA and diphtheria toxin.

A patient was suspected with diphtheria if he/she presented at least 2 of the following symptoms: fever, sore throat, reddened throat, cervical lymph node swelling, difficulty breathing, throat irritation, cough, swallowing difficulties, or a whitish or grayish throat membrane. An epidemiologically linked case corresponded to the definition of a suspected case that had been in contact with a laboratory-confirmed case within 14 days preceding the onset of disease signs. A clinically compatible case was defined as any case meeting the definition of a suspected case with no laboratory confirmation test results and no epidemiological link with a laboratory-confirmed case. A discarded case was any suspected case with 2 RT-qPCR tests that yielded negative results for the diphtheria toxin gene (toxA and toxB) [13, 14].

Laboratory Analysis

The nasopharyngeal samples were collected by a trained biologist. The collection kit included a nasopharyngeal swab, the AMIES transport medium (Thermo Fisher Scientific), and personal protective equipment [15]. These samples were securely packaged in triple layers and maintained at a temperature of 4°C–8°C. Further, they were classified using data from the investigation forms, and transported to the National Public Health Laboratory and then to the CERFIG for diagnostic confirmation.

Manual extraction of bacterial DNA was performed using a human diagnostic kit (RunMei Gene Technology Co., Ltd.), according to the manufacturer's instructions. Fresh DNA extracts were used to confirm the diagnosis via RT-qPCR. The DNA aliquots were subsequently stored at −20°C.

The confirmation of diphtheria diagnosis was based on the presence of diphtheria toxins in suspected patients. The diphtheria diagnosis was biologically confirmed after establishing an epidemiological link and a clinical diagnosis of the disease at the epidemiological treatment centers of the reporting districts of the cases. This strategy refers to the recommendations of the Pasteur Institute, which involves confirming the clinical diagnosis via a test for detecting the toxin secreted by *C. diphtheriae* [16]. The primer and probe sequences used to detect the 2 subunits (A and B) of the diphtheria toxin were as follows: ToxA-forward—GGCGTGGTCAAAGTGACGTA; ToxA-reverse—CTTGCTCCATCAACGGTTCA; ToxA-probe—6FAM-CCAGGACTGACGAAGGTTCTCGCACT; ToxB-forward—CG CCCTAAATCTCCTGTTTATGTT; ToxB-reverse—GTACCCAA GAACGCCTATGGAA; and ToxB-probe—6FAM-TTCACAGAA GCAGCTCGGAGAAAATTCATTC [17].

The primers were reconstituted at a concentration of 20 μM and the probes at 5 μM. To prepare the real-time qPCR mixture, we used 10 µl of the Master Mixes using the LightCycler® 480 RT-qPCR system, 0.5 µl of each primer, 0.5 µl of the probe, 3.5 µl of H_2O, and 5 µl of DNA extract. The amplification program on the CFX 96 real-time PCR system (Bio-Rad) was as follows: 1 cycle at 95°C for 10 min, followed by 40 cycles of 95°C for 10 s and 60°C for 30 s and finally by 1 cycle at 40°C for 30 s [17]. Each RT-qPCR plate contained the 2 pairs of primers accompanied by the corresponding probes and allowed the simultaneous detection of the 2 subunits of the diphtheria toxin. The validation criteria for the RT-qPCR results included a Ct value of >38, a sinusoidal amplification curve for the positive control, and lack of amplification in the negative controls.

Data Analysis

A descriptive analysis of the data was performed. Qualitative variables were expressed as frequencies, and quantitative variables were presented as means with standard deviation. A bivariate analysis using the χ^2 test and the Fisher's exact test was performed to investigate the association between diphtheria positivity and the sociodemographic, clinical, and exposure characteristics of the participants. Next, variables with a *P*-value of <.20 were included in a multivariate logistic regression model to identify factors that are independently associated with diphtheria positivity. Odds ratios and confidence intervals were then calculated, and a *P*-value of <.05 indicated statistically significant differences. Data were entered into the Excel sheet and analyzed using the R-Studio 4.4.1 software.

Patient Consent Statement

This study was conducted in the context of a public health emergency related to the diphtheria epidemic in the country. Consequently, the inclusion of participants was opportunistic and based on routine care. Therefore, explicit consent was not required. The patients were informed that their sociodemographic and clinical information and blood samples would be used to coordinate the epidemic response. The National Ethics Committee for Health Research (CNERS) of Guinea approved the study protocol under the reference number N°186/CNERS/23 dated October 24, 2023.

RESULTS

Diphtheria Positivity Rate

Over a 1-year epidemic period, we received 444 samples collected from patients suspected of diphtheria, of whom 90 tested positive on 2 RT-qPCR tests, resulting in a global positivity rate of 20.3%. In total, 13 deaths, including 8 confirmed cases, were recorded, yielding a case fatality rate of 8.9% (Figure 1).

Sociodemographic Characteristics of the Participants

The median age of the patients with diphtheria was 11 ± 9 years (min: 1 year, max: 60 years), with a female predominance (53 [59%] of 90 patients) and a male-to-female sex ratio of 0.7. Among all patients who tested positive, only 3 (3.3%) were vaccinated against diphtheria. Of them, only 1 had a vaccination card. In total, 53 (59%) of 90 patients with diphtheria had been in contact with a patient during disease onset. Meanwhile, 27 (30%) of 90 patients reported no contact with a sick person, and 10 (11%) of 90 patients were not aware if they had prior contact (Table 1).

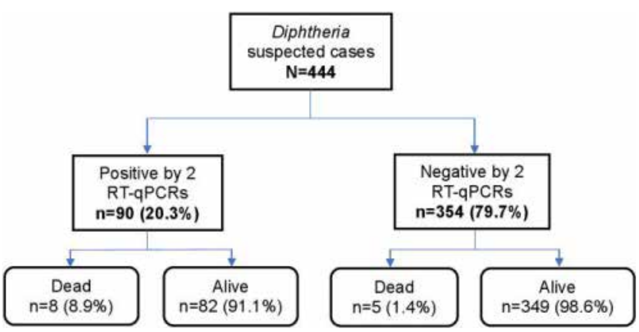

Figure 1. Flowchart for the selection of diphtheria cases. We have collected 444 samples from patients suspected of diphtheria. Among these patients, 90 tested positive for diphtheria, and 354 tested negative. Among the diphtheria cases, 8 deaths and 82 recoveries were recorded. In addition, 5 deaths were reported among the patients who tested negative.

Table 1. Sociodemographic Characteristics, Risk Factors, and Clinical Signs of the 444 Patients Suspected of Diphtheria Who Underwent Confirmation Testing

Characteristics of the Patients	Patients Who Were Diphtheria-negative Based on the 2 RT-qPCR Tests *N* = 354[a]	Patients Who Were Diphtheria-positive Based on the 2 RT-qPCR Tests *N* = 90[a]	*P*-Value[b]
Age (y)	…	…	<.001
1–5	85 (24%)	53 (59%)	…
6–10	89 (25%)	17 (19%)	…
11–15	59 (17%)	11 (12%)	…
> 15	121 (34%)	9 (10%)	…
Sex	…	…	.6
Female	218 (62%)	53 (59%)	…
Male	136 (38%)	37 (41%)	…
Region	…	…	.002
Conakry	45 (13%)	8 (8.9%)	…
Faranah	15 (4.2%)	3 (3.3%)	…
Kankan*	209 (59%)	72 (80%)	…
Kindia	13 (3.7%)	3 (3.3%)	…
Labé	9 (2.5%)	0 (0%)	…
Mamou	63 (18%)	4 (4.4%)	…
Vaccinated patients**	41 (12%)	3 (3.3%)	.019
Patients with vaccination records	15 (4.2%)	1 (1.1%)	.2
Contact with a patient	129 (36%)	53 (59%)	<.001
No contact with a patient	213 (60%)	27 (30%)	…
Lack of awareness about contact with a patient	12 (3.4%)	10 (11%)	…
Clinical symptoms	…	…	…
Fever	314 (89%)	81 (90%)	.7
Headaches	214 (60%)	51 (57%)	.5
Cough	169 (48%)	51 (57%)	.13
Dyspnea	45 (13%)	25 (28%)	<.001
Physical asthenia	93 (26%)	32 (36%)	.080
Abdominal pain	4 (1.1%)	1 (1.1%)	>.9
Nausea	68 (19%)	16 (18%)	.8
Vomiting	55 (16%)	15 (17%)	.8
Sore throat	307 (87%)	82 (91%)	.3
Dysphagia	292 (82%)	80 (89%)	.14
Swollen neck lymph	103 (29%)	33 (37%)	.2
Throat irritation	109 (31%)	27 (30%)	.9
Throat redness	266 (75%)	73 (81%)	.2
Whitish throat membrane	224 (63%)	75 (83%)	<.001
Skin lesion	6 (1.7%)	3 (3.3%)	.4
Joint damage	2 (0.6%)	1 (1.1%)	.5
Dysphonia	2 (0.6%)	0 (0%)	>.9
Rhinorrhea	7 (2.0%)	1 (1.1%)	>.9

The variables associated with diphtheria were age (*P*-value < .001), health region or endemic area (*P*-value < .002), contact with a patient (*P*-value < .001), dyspnea (*P*-value < .001), and whitish throat membrane (*P*-value < .001).

[a] *N* (%).

[b] Pearson's χ^2 test; Fisher's exact test.

* Siguiri district where all 8 fatal cases occurred is a part of this region.

** The vaccination against diphtheria had been reported by the patient's companion or the patient him/herself.

Six of the 8 administrative regions recorded suspected cases of diphtheria. Of these, 5 had confirmed cases. Among the affected regions, the Kankan region had the highest number of diphtheria cases (80%, 72 of 90 patients). In addition, the Conakry, Mamou, Faranah, and Kindia regions recorded proportions of confirmed cases at 8.9% ($n = 8$), 4.4% ($n = 4$), and 3.3% ($n = 3$), respectively. However, the Labé region did not record any confirmed cases of diphtheria despite reporting 9 suspected cases (Table 1).

Clinical Signs and Symptoms

The common clinical characteristics of patients with diphtheria were sore throat (91%), fever (90%), dysphagia (89%), whitish throat membrane (83%), redness at the back of the throat (81%), headaches and cough (57%), physical asthenia (36%), cervical lymph node swelling (37%), throat irritation (30%), and dyspnea (28%) (Table 1).

Eight Positive Cases of Diphtheria based on 2 RT-qPCR Tests

The 8 diphtheria-related deaths, which involved 6 girls and 2 boys, with a median age of 4 years, occurred in the Siguiri district. Upon admission to the epidemic treatment center, the predominant clinical features of the patients who died were as follows: fever ($n = 8$), generalized asthenia ($n = 8$), odynophagia ($n = 8$), dysphagia ($n = 8$), presence of diphtheritic membrane ($n = 8$), throat redness ($n = 6$), cervical edema ($n = 6$), and cough ($n = 6$). None of the patients had received diphtheria vaccination. On average, the patients died 2 days after admission. Six of 8 patients (4 girls and 2 boys) died within the first 3 months after confirming the epidemic. Meanwhile, only 1 death was recorded in both the second and fourth quarters of the year.

Weekly Trends in Suspected, Confirmed, and Negative Cases of Diphtheria

The positivity rates were 30.0% (9 of 30 patients) during the first quarter (from July to September 2023), 19.3% (26 of 135 patients) during the second quarter (from October to December 2023), 3.7% (5 of 135 patients) during the third quarter, and 34.7% (50 of 144 patients) during the fourth quarter (Figures 2*A* and 2*B*).

Risk Factors

The risk factors associated with diphtheria included age <15 years, with the specific details as follows: 1–5 years (adjusted odds ratio [aOR] = 10.6, 95% confidence interval [CI] = 4.83–25.4), 6–10 years (aOR = 3.38, 95% CI = 1.41–8.59), and 11–15 years (aOR = 2.84, 95% CI = 1.08–7.68). Lack of vaccination against diphtheria (aOR = 3.76, 95% CI = 1.21–16.7) and contact with a patient with confirmed diphtheria (aOR = 3.20, 95% CI = 1.83–5.71) were also a significant risk factor. The predictive signs of diphtheria included a whitish throat membrane (aOR = 4.10, 95% CI = 2.13–8.42) and dyspnea (aOR = 2.68, 95% CI = 1.27–5.65) (Table 2).

DISCUSSION

The Republic of Guinea has become a hotspot for epidemic-prone diseases over the last decade. Since 2023, the country has been facing a diphtheria outbreak.

It should be emphasized that the diphtheria vaccine has been introduced in Guinea in the 1980s as part of the World Health Organization's Expanded Program on Immunization. Thereafter, this program has ensured the implementation of the country's vaccination strategy. The vaccination coverage remains a challenge, with rates below 50% since 2014 [18].

In 2020, administrative data reported that ∼90% of infants aged 0–11 months received their third dose of the pentavalent vaccine in Guinea. However, the full vaccination coverage rate for children aged 12–23 months was only 24% [19]. This is still insufficient to prevent the occurrence of diphtheria in the country. Several studies have shown that vaccination alone can only interrupt transmission in 28% of epidemic households [20, 21]. Countries affected by diphtheria have implemented strategies to strengthen routine vaccination, organize catch-up campaigns, enhance surveillance and early detection, train healthcare personnel, improve technical capacities, and raise community awareness about best practices while promoting intersectoral cooperation [22]. Thus, the implementation of these strategies will significantly enhance vaccination coverage and, as a result, can effectively control the diphtheria epidemic.

In our case series, diagnosis was based on clear epidemiological link, compatible clinical presentation, and 2 RT-qPCR tests that detected the diphtheria toxin gene in blood samples [15]. However, bacterial culture and the Elek test, which are the gold standards for confirming diphtheria, could not be performed because of technical difficulties.

Data were collected from individuals suspected of diphtheria. Each patient was identified using a notification form filled out based on the declarations of the patients themselves or their companions. The positivity rate was 20%, which is one of the highest in the West African subregion. According to an AFRICA-CDC report, the diphtheria epidemic has affected 4 African countries including Algeria, Guinea, Niger, and Nigeria. As of 9 October 2023, there have been over 14 587 reported cases, with a case fatality rate of 4.1%. Nigeria accounts for >90% of these cases [23]. This high positivity rate could be explained by the screening strategy implemented by the country's health authorities. This strategy was based on the detection of new epidemic foci, the hospital management of positive cases, and the vaccination of contacts aged >5 years.

After confirming the initial diphtheria cases in Siguiri, a city in the Kankan region, the disease spread to 5 other regions along National Highway N° 01, likely caused by patient or contact mobility. This underscores the need for a comprehensive contact investigation to identify and manage affected individuals, especially due to the complexity added by multiple

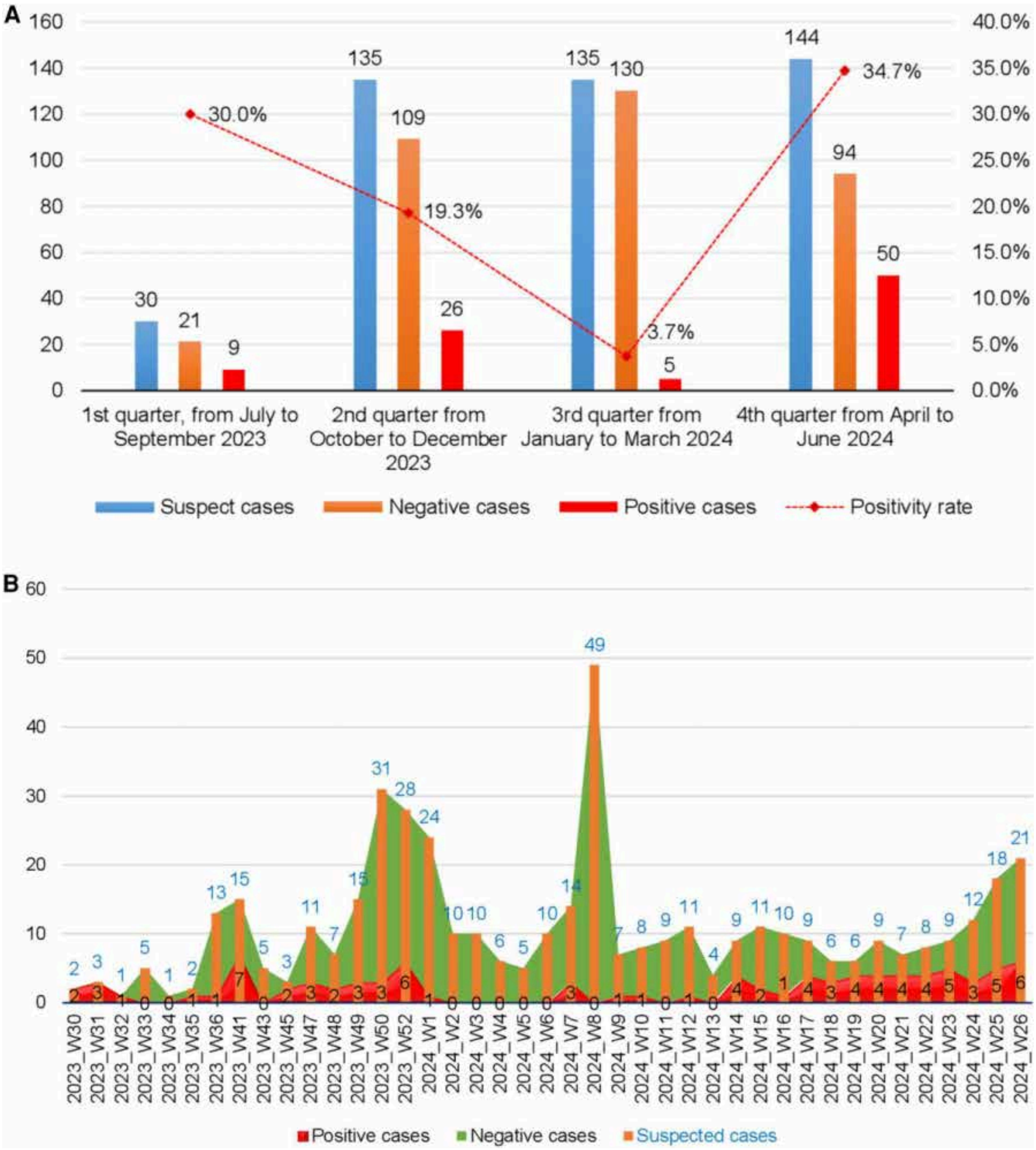

Figure 2. *A* and *B*, Weekly and quarterly evolution of diphtheria cases from July 2023 to June 2024 in the Republic of Guinea. During the first quarter of the epidemic, 30 samples from suspected individuals were analyzed, with a total of 9 confirmed cases of *diphtheria*, which corresponded to a positivity rate of 30%. In the second and third quarters, 135 samples collected from suspected individuals were analyzed, with 26 and 5 confirmed cases, yielding positivity rates of 19.3% and 3.7%, respectively. During the fourth quarter, 144 samples obtained from suspected individuals were tested. Among the patients, 50 were confirmed to be positive, leading to a positivity rate of 34.7%.

epidemic foci. Rapid, resource-intensive responses are essential to address this urgent health challenge.

The confirmed cases peaked at 2 times points throughout the year: October–December 2023 (29%) and April–June 2024 (55%). Despite efforts, these peaks emphasize the continued public health threat caused by diphtheria, exacerbated by new epidemic foci and disease spread within communities. These periods follow the rainy season (July–September) and the dry season (January–March). Thus, climatic factors may influence future epidemic management. However, a direct link has not been established.

Overall, this study showed that children aged 1–5 years were the most affected, with a predominance of girls. There were only 3 patients who were confirmed to have been initially vaccinated, and only 1 had a vaccination card. All 3 survived. In 2023 in Nigeria, 73.5% ($n = 3466$) of patients with confirmed

Table 2. Factors Associated With Diphtheria

Characteristics of the Patients	OR[a]	95% CI[a]	P-Value
Age (y)	Adjusted value	…	
1–5	10.6	4.83–25.4	< .001
6–10	3.38	1.41–8.59	.008
11–15	2.84	1.08–7.68	.035
> 15	1.0	…	
Patients vaccinated against diphtheria	1.0	…	
Patients not vaccinated against diphtheria	3.76	1.21–16.7	.041
No contact with a patient	1.0	…	
Contact with a patient with diphtheria	3.20	1.83–5.71	< .001
Lack of awareness about contact with a patient	2.86	0.95–8.49	.058
Dyspnea	2.68	1.27–5.65	.010
Whitish throat membrane	4.10	2.13–8.42	< .001

Compared with patients aged 15 y, those aged 1–5 y were 10.6 times (95% CI: 4.83–25.4) more likely to develop diphtheria. Children aged 6–10 y were 3.38 times (95% CI: 1.41–8.59) more likely to contract the disease. Meanwhile, children aged 11–15 y were 2.84 times (95% CI: 1.08–7.68) more likely to contract diphtheria. Unvaccinated children (adjusted OR: 3.76, 95% CI: 1.21–16.7) were significantly more likely to contract diphtheria, similar to those who had been in contact with an infected person (adjusted OR: 3.20, 95% CI: 1.83–5.71). Dyspnea (adjusted OR: 2.68, 95% CI: 1.27–5.65) and a whitish throat membrane (adjusted OR: 4.10, 95% CI: 2.13–8.42) were significantly associated with diphtheria.

[a]OR, odds ratio; CI, confidence interval.

diphtheria were aged 1–14 years, with a female predominance of 56.3%. Only 22.8% ($n = 1074$) were fully vaccinated against diphtheria, and 6% were partially vaccinated [24]. In India, based on a laboratory-supported diphtheria case surveillance, a female predominance of 69% was observed in 2016 [25]. In addition to the predominance of children, 10% of positive diphtheria cases involved adults, as observed in other studies [13, 26–28]. This finding confirms that diphtheria continues to pose a threat to both children and unvaccinated or under-immunized adults, thereby emphasizing the ongoing importance of vaccination and vigilance. Notably, 1 patient who received the DTP-Hib-HepB (pentavalent) vaccine tested positive for diphtheria, thereby raising concerns about the immunity levels in vaccinated populations. This underscores the need to evaluate antibody levels in vulnerable groups, particularly during epidemics [29]. Regarding the higher number of women who tested positive for diphtheria, no biological or immunological differences were noted between sexes thus far.

The analyses also revealed that lack of vaccination and contact with infected individuals are significant risk factors for the outcome. In addition, the presence of dyspnea and diphtheritic membrane is strongly associated with the outcome. In any case, several factors must be considered in the clinical symptomatology of diphtheria. These results underscore the need to strengthen diphtheria vaccination, particularly among children aged 1–15. They also highlight the importance of training healthcare workers to immediately recognize the signs of the disease, to facilitate early diagnosis and treatment, and to effectively manage contact cases to limit transmission.

CONCLUSION

Although diphtheria can be prevented by vaccination, it has resurfaced again in Guinea, with a high fatality rate. Children are generally affected. The main clinical signs of the disease are sore throat, fever, dysphagia, and the presence of a whitish membrane observed against a reddened, inflamed background. The risk factors associated with diphtheria include age <15 years, lack of immunization, and contact with an infected individual. The presence of a whitish membrane at the back of the throat and dyspnea are the predictors of the disease. This study emphasized the importance of vaccination and the strengthening of programs, along with early diagnosis and treatment, in inhibiting the impact of diphtheria. Blood culture and the Elek test remain important for diagnostic confirmation. However, the detection of diphtheria toxin via RT-qPCR has been confirmed to be essential for accelerating epidemic response, particularly in challenging cases.

Notably, in addition to its socio-health impact, this re-emergence reflects the fragility of the whole health system in the country, the failure of vaccination strategies, and the challenge of eliminating vaccine-preventable diseases. This study addresses a gap in the research of epidemic-prone diseases in general and particularly diphtheria. The analyzed data encompass nearly all health regions across the national territory. We believe that the results obtained are sufficient to describe the extent of the epidemic, draw lessons from it, and develop methods that can improve the coordination of our response to diphtheria.

Notes

Acknowledgments. We would like to extend our gratitude to the management of CERFIG and the team at the molecular biology laboratory for their dedication to this work. We are also thankful to our colleagues at the Hospital-University Institute of Aix-Marseille University for supplying some of the reagents essential for these analyses. Further, we appreciate the support of the national health authorities, particularly the leaders of the epidemiological treatment centers, who provided us with samples from patients suspected of diphtheria.

Author Contributions. A. K. K., A. T., F. F., O. M., and A. K. K. designed the study. A. K. K., A. K. S., A. K., M. C., H. D., and J. B. K. conducted the confirmation tests at the CERFIG Laboratory. A. K. K. collected data from the patient notification forms and created the study database. A. K. K., T. A. C. G., S. T. B., and K. J. J. O. K. analyzed the data. A. K. K. wrote the first version of the article. A. T., F. F., O. M., and A. K. K. supervised and coordinated all the data analyses and the drafting of the manuscript. All authors have read and approved the final version of the article.

Financial support. This work was carried out at CERFIG (Centre de Recheche et de Formation en Infectiologie de Guinée) with funding from the AfriCam Project of the PREZODE (Preventing ZOnotic Diseases Emergence) Initiative, as part of its support for the Guinean Ministry of Health and Public Hygiene in response to epidemics and surveillance of diseases with epidemic potential. This work was also supported by a grant from the French Government managed by the National Research Agency under the "Investissements d'avenir (Investments for the Future)" program with reference number ANR-10-IAHU-03 (Méditerranée Infection), by the

Contrat Plan Etat-Région and the European funding FEDER IHUPERF. This work was carried out with the support of the laboratories and research units in which the doctoral student carries out his research. These various bodies receive funding from the donors mentioned. It is therefore not a research project financed exclusively by a Sponsor, but rather the technical, material and financial support of a student.

Potential conflicts of interest. There are no conflicts of interest to disclose. The research institutions and researchers involved in this work have no evident or hidden financial interests. The authors: No reported conflicts of interest. All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest.

References

1. Chaudhary A, Pandey S. Corynebacterium Diphtheriae, eds. Statpearls. Treasure Island (FL): StatPearls Publishing, **2023**. Available at : http://www.ncbi.nlm.nih.gov/books/NBK559015/
2. Par Estelle B. Diphtérie : définition, épidémiologie, symptômes, diagnostic. Santé sur le Net, l'information médicale au cœur de votre santé. 2019. Available at: https://www.sante-sur-le-net.com/maladies/maladies-infectieuses/diphterie/
3. Ventura M, Canchaya C, Tauch A, et al. Genomics of actinobacteria: tracing the evolutionary history of an ancient phylum. Microbiol Mol Biol Rev **2007**; 71: 495–548.
4. Sharma NC, Efstratiou A, Mokrousov I, et al. Diphtheria. Nat Rev Primer **2019**; 5: 81.
5. Antunes CA, Sanches dos Santos L, Hacker E, et al. Characterization of DIP0733, a multi-functional virulence factor of Corynebacterium diphtheriae. Microbiol Read Engl **2015**; 161:639–47.
6. Larry MB, Maria TVP. Édition professionnelle du Manuel MSD. 2023. Diphtérie—Maladies infectieuses. Available at: https://www.msdmanuals.com/fr/professional/maladies-infectieuses/bacilles-gram-positifs/dipht%C3%A9rie
7. Nguyen Thi Nguyen T, Parry CM, Campbell JI, et al. Endemic erythromycin resistant corynebacterium diphtheriae in Vietnam in the 1990s. Microb Genomics **2022**; 8:mgen000861.
8. Beaudet MF, Poitras A. Diphtérie: hausse des cas dans plusieurs régions du monde. Inst Natl Santé Publique Qué **2022**; 22. https://www.inspq.qc.ca/sites/default/files/bulletins/actu_2022_vol_23_num_11.pdf
9. Organisation Mondiale de la Santé. Diphtérie, principaux faits en 2022. 2023. Available at: https://www.who.int/fr/news-room/fact-sheets/detail/diphtheria
10. Ikejezie J, Adebusoye B, Ekezie W, et al. Modifiable risk factors for diphtheria: a systematic review and meta-analysis. Glob Epidemiol **2023**; 5:100100.
11. ANSS—Agence National de Sécurité Sanitaire. SitRep N°303 du 16 Novembre 2024. Available at: https://anss-guinee.org/
12. Institut national de la Statistique. Enquête Démographique et de Santé (EDSG V—2018), Indicateurs-Clés. République de Guinée: Ministère du Plan et du Développement Economique; 2018. Available at: https://www.stat-guinee.org/images/Documents/Publications/INS/rapports_enquetes/EDS-2018-Indicateurs-Cles.pdf
13. Polonsky JA, Ivey M, Mazhar MDKA, et al. Epidemiological, clinical, and public health response characteristics of a large outbreak of diphtheria among the Rohingya population in Cox's Bazar, Bangladesh, 2017 to 2019: a retrospective study. PLoS Med **2021**; 18:e1003587.
14. Strauss RA, Herrera-Leon L, Guillén AC, et al. Molecular and epidemiologic characterization of the diphtheria outbreak in Venezuela. Sci Rep **2021**; 11:6378.
15. Word Health Organization, Androulla E, Fatima S, et al. Diagnosis of diphtheria and other related infections, third revision. Available at: https://cdn.who.int/media/docs/default-source/immunization/diphtheria_lab_manual_v2.pdf?sfvrsn=3e1d6f8f_3
16. Institut Pasteur 2015. Comment diagnostiquer la Diphtérie. Available at: https://www.pasteur.fr/fr/centre-medical/fiches-maladies/diphterie
17. Mothershed EA, Cassiday PK, Pierson K, et al. Development of a real-time fluorescence PCR assay for rapid detection of the diphtheria toxin gene. J Clin Microbiol **2002**; 40:4713–9.
18. Organisation Mondiale de la Santé. Diphtérie—Guinée. 2023. Available at: https://www.who.int/fr/emergencies/disease-outbreak-news/item/2023-DON492
19. Guinea-2020-COAR. Available at: https://www.unicef.org/media/100236/file/Guinea-2020-COAR.pdf
20. Truelove SA, Keegan LT, Moss WJ, et al. Clinical and epidemiological aspects of diphtheria: a systematic review and pooled analysis. Clin Infect Dis **2020**; 71: 89–97.
21. Ogunniyi TJ. Revealing the challenges and efforts of routine immunization coverage in Nigeria. Health Sci Rep **2024**; 7:e70000.
22. Strategic Partnerships Towards Transforming Health Systems and Outcomes in Nigeria. Strategic partnerships towards transforming health systems and outcomes in Nigeria. | WHO | Regional Office for Africa. Available at: https://www.afro.who.int/countries/nigeria/news/strategic-partnerships-towards-transforming-health-systems-and-outcomes-nigeriastrategic
23. Africa CDC. Diphtheria outbreak in Africa: strengthening response capacities. Africa CDC. Available at: https://africacdc.org/news-item/diphtheria-outbreak-in-africa-strengthening-response-capacities/
24. Oduoye MO, Marsool MDM, Haider MU, et al. Unmasking diphtheria in Nigeria: a multifaceted approach to tackle outbreaks and improve immunization rates among the Nigerian population—an updated correspondence. Health Sci Rep **2024**; 7:e1804.
25. Sangal V, Hoskisson PA. Evolution, epidemiology and diversity of Corynebacterium diphtheriae: new perspectives on an old foe. Infect Genet Evol **2016**; 43:364–70.
26. Moghalles SA, Aboasba BA, Alamad MA, et al. Epidemiology of diphtheria in Yemen, 2017–2018: surveillance data analysis. JMIR Public Health Surveill **2021**; 7:e27590.
27. Besa NC, Coldiron ME, Bakri A, et al. Diphtheria outbreak with high mortality in northeastern Nigeria. Epidemiol Infect **2014**; 142:797–802.
28. Sangal L, Joshi S, Anandan S, et al. Resurgence of diphtheria in North Kerala, India, 2016: laboratory supported case-based surveillance outcomes. Front Public Health **2017**; 5:218.
29. Mosnier A, Daviaud I, Clairaz B, et al. Nouvelle stratégie de vaccination de rappel dTcaPolio chez l'adulte: qu'en pensent les vaccinateurs? Médecine Mal Infect Form **2023**; 2(2, Supplement):S142–3.