Exploring the Larvicidal Potential of Schiff Bases and Their Cobalt Complexes: New perspective on Malaria Vector Control

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Abstract: Vector-borne illnesses including dengue, yellow fever, and malaria present serious health threats, particularly in sub-Saharan Africa. Malaria is still a major cause of morbidity and mortality, innovative strategies for vector control, are required, particularly those that target the immature stages of mosquitoes. The resistanceto current antimalarial treatments makes the development of new insecticides targeted at these larval stages essential. The goal of this research is to create chemical compounds, namely Schiff bases that have the potential to be efficient insecticides against the vectors of malaria. Two Schiff bases namely 5- nitrosalicylidene-2-aminopyridine (L1) and 5-bromosalicylidene-2-aminopyridine (L2) were synthesized *via* condensation of 2- aminopyridine and salicylaldehyde derivatives. Their cobalt complexes (CoL1 and CoL2) were prepared from cobalt (II) chloride hexahydrate in water. The compounds were characterized through melting point, elemental analysis, infrared (IR) spectroscopy, nuclear magnetic resonance (NMR) spectroscopy, and electronic spectra. The susceptibility of *Anopheles gambiae* larvae to the synthesized compounds was assessed using a methodology adapted from the Elliot larval test. The larval bioassay was conducted in different concentrations from 250µl/100ml - 0.5 µl/100ml in duplicates using 25 mosquitoes of both field andlaboratory strains in each exposure for 24, 48 and 72 hours respectively. Data obtained from the spectroscopic and analytical techniques supported the structures proposed for the synthesized compounds. Allcompounds exhibited similar activity at 250-125 µl/100ml against the laboratory strain. However, CoL2-the bromo-containing complex exhibited highest activity at 31.25 µl/100ml against both strains. The 72-hour assay also revealed that bromo -containing compounds (L2 and CoL2) displayed higher level of toxicity against the strains compared to the nitro-containing compounds (L1 and CoL1) at 250 -65.5µl/100ml. The study highlights the significance of creating new insecticides that target the larval stages of malaria vectors, paving the way for more efficient control strategies against this persistent public health threat.

Keywords: Anopheles gambiae, cobalt (II) complex, insecticides, larvicidal, Schiff base

Introduction

Pathogens spread by insects and other arthropods vectors among humans, animals, or plants have caused a great deal of morbidity and mortality. Vector-borne diseases such as dengue, malaria, plaque and yellow fever, collectively caused more illness and fatalities in humans than all other causes combined (Nicholson *et al*., 2019). Of a significant public health concern in Africa sub-Saharan (Nigeria) is malaria (CDC, 2021, USAID, 2022). The *Plasmodium* parasite, a single-celled parasite that is spread to persons by the bite of *Anopheles* mosquitoes, is the source of this disease, which is the most enduring parasitic illness affecting humans (Dondorp *et al*., 2010, Belete, 2020). The growth of antimalarial agents to the eradication of malaria parasites has experienced scientifically significant advancements. This covers, but is not restricted to chloroquine, artemisinin-based combination therapy (ACT)and quinine to treat infected patients (WHO, 2014). Nevertheless, numerous malaria-related deaths are still reported each year all over the world due to the parasite resistance that is evolving (Belete, 2020, Foster & Walker, 2009).

It is therefore imperative to put in place control measures tocurb the disease burden. According to WHO, the primary strategy for stopping the spread of malaria and preventing its vector, (mosquito), is vector control (WHO, 2021). A crucial component in the fight against malaria is vector control and elimination methods, given that it's a highly effective strategies to decrease the spread of malaria. In addition to the elimination of malaria vectors in the majority of places at risk, the World Health Organization currently recommends the use of insecticide-treated nets (ITNs) or indoor residual spraying (IRS) (WHO, 2024b).

The Use of insecticides has been the most popular approach among these vector control strategies. Most insecticides if not all that make up over 25% of the global pesticide market contain pyrethroids as the most active ingredient (Scott *et al*., 2015). In Africa, pyrethroids are the sole thatis advised for treating mosquito nets in order to avoid malaria. However, pesticide resistance that has developed in mosquito vectors and parasite drug resistance are the main reasons why mosquito-borne diseases are currently reemerging (Liu *et a*l., 2006). The mosquito control effort is currently facing significant challenges due to the widespread resistance to insecticides, particularly pyrethroid resistance, and the fact that resistance to one pesticide typically results in cross resistance to other insecticides (Zaim & Guillet, 2002).

According to Hoover (2005), the application of

pesticidesis widely acknowledged as the least effective strategy of a sustainable reduction in mosquito populations. This isattributed to pesticide resistance, pest/vector resurgence,secondary pest outbreaks, death of non-target organisms, environmental contamination/pollution, bioaccumulation potentials in the trophic structure of ecosystems (Hoover, 2005). It is therefore critically necessary to create innovative substitutes and/or additional control methods in order to prevent diseases spread by mosquitoes.

One of the main alternative strategies of effectively reducing the disease burden is through the control of immature stages of vector populations, which are restricted to aquatic environment. Out of the four stages of mosquito life cycle, all the immature stages are confined to the aquatic medium, thus providing an opportunity for the effective control of these insects just before the adults resume their role as disease transmitters. The control of larval stages of mosquitoes is referred to as larviciding, and the killing agents called larvicides. Larviciding has the ability to solve a number of the problems that malaria vector management is now confronting (Choi & Wilson, 2017). Firstly, larviciding can target mosquitoes that bite and rest outdoors and are less susceptible to the effects of IRS and LLINs (WHO, 2024b). Second, if high coverage ofLLINs and IRS is insufficient to eradicate malaria, it might be utilized to address residual foci of the disease. Finally, larviciding (Cui *et al*., 2006, WHO, 2024a) could be employed as a component of an insecticide resistance management strategy in conjunction with otherinterventions.

It is thus vitally necessary to create unique insecticides of new chemical compounds that focus on larval stages of mosquito. In addition to other interventions, compounds containing Schiff bases (Sharma *et al*., 2014, Chandramouli *et al*., 2012) as a structurally essential component are a useful research tool for investigating novel larvicides (Ali*et al*., 2009) due to their ease of synthesis, environmental friendliness and unique anti-malarial activity which has been reported to improve upon complexion with transition metal ions (Joshi *et al*, 2022). These compounds could be used as part of an insecticide resistance management strategy. This advancement will improve human health and wellbeing (SDG 3) (Frank *et al*., 2017). In continuation of this work, it is reported herein the acute toxicity of 2-aminopyridine based Schiff bases and their corresponding cobalt (II) complexes on larvae of *Anopheles gambiae* in laboratory bioassays.

Materials and Method

Chemicals

2-Aminopyridine, 5-bromo salicyladehyde, 5-nitro salicyladehyde and cobalt(II) hexahydrate were purchased from Aldrich Chemicals Co. Ltd. Ethanol, hexane, and dimethyl sulfoxide (DMSO) also came from Aldrich Chemicals and were utilized without additionalpurification; they were of analytical or spectroscopic grade.

Methods

Thin-layer chromatography (TLC) plate's pre coated with silica gel 60 F254 were used for TLC under the UV wavelengths of 365 nm and 264 nm. Melting point tests were conducted using a melting point apparatus (Stuart SMP3), and the results remained consistent. Following the user manual's instructions, infrared spectrometer of a FTS 7000 series Digilab Win-IR Pro fitted with an attenuated total reflectance (ATR) Diamond Selenium attachment wasused. The resulting spectra fell between 4000 and 400 cm^{-1}.When using nuclear magnetic resonance (NMR) spectroscopy for carbon (^{13}C) and proton (^{1}H), deuterated chloroform ($CDCl_3$) was employed as the solvent, with TMS (Tetramethylsilane, internal standard) is utilized. Chemical shifts were measured at room temperature and compared to the solvent peaks. The CHN/M analysis was done on a Perkinelmer 2400 CHNS/O analyzer. Following

solution preparation in DMF, with a 1 cm quartz cell connected to a Cecil Super Aquarius 9000 series UV-Vis spectrophotometer, the electronic spectra of the compoundswere captured at ambient temperature.

Typical Procedure for the synthesis of Schiff bases (L1-L2)

N-(5-nitro-salicylidene)-2-aminopyridine (L1)

In a 10 mL of hot absolute ethanol, was added 0.01 mmol (16.70 mg) of 5-nitro-salicylaldehyde. This was mixed with2-aminopyridine (0.10 mmol, 9.40 mg) in 5 mL of hotabsolute ethanol. The combination used in the reaction was refluxed for eight hours at 65 °C. A yellow crystals appeared in the mixture as it cooled to room temperature, and these were subsequently collected by filtering. The crystals were purified using an ethanol: hexane (1:1) to yield pure **L1**. A yield of (46%) 11.05 mg was attained for the final product. Melting point: 182 to 184 °C; Rf: 0.68.

Characterization Data:

- **IR (cm^{-1}):** 3331, 1595, 1556, 1526, 1426, 1373, 1285, 1195, 1147, 1099, 1023, 930, 863, 728, 646, 552
- **^{1}H NMR (300 MHz, $CDCl_3$):** δH: 14.56 (s, 1H),9.53 (s, 1H), 7.10–8.54 (m, 7H),
- **^{13}C NMR (75 MHz, $CDCl_3$):** δC: 167.63 (C-OH), 162.89 (HC=N),

Analytical Data:

$C_{12}H_9N_3O_3$ was calculated (found) as follows: H, 3.73 (3.56); C, 59.26 (59.14); N, 17.28 (16.96)

N-(5-bromo-salicylidene)-2-aminopyridine (L2)

In a 10 mL of hot absolute ethanol, was added 0.01 mmol (20.10 mg) of 5-bromo-salicylaldehyde. This was mixed with 2-aminopyridine (0.10 mmol, 9.40 mg) in 5 mL of hotabsolute ethanol. The combination used in the reaction wasrefluxed for six hours at 60 °C. Light orange crystals appeared in the mixture as it cooled to room temperature, and these were subsequently collected by filtering. The crystals were purified using an ethanol: hexane (1:1) to yield pure L2. A yield of (81%) 22.70 mg was attained for the final product. Melting point: 138 to 140 °C; Rf: 0.50.

Characterization Data:

- **IR (cm^{-1}):** 1608, 1586, 1550, 1461, 1428, 1381, 1341, 1276, 1184, 1144, 1100, 990, 918, 870, 814, 782, 700, 628, 607
- **^{1}H NMR (300 MHz, $CDCl_3$):** δH: 13.42 (s, 1H),9.34 (s, 1H), 6.89–8.49 (m, 7H),
- **^{13}C NMR (75 MHz, $CDCl_3$):** δC: 163.40 (C-

OH), 160.86 (HC=N),

Analytical Data:

$C_{12}H_9N_2OBr$ was calculated(found) as follows: H, 3.27(3.21); C, 52.01(51.96) ; N, 10.10 (9.88).

Synthesis of Cobalt (II) complexes of the Schiff bases

Cobalt (II) Complex of L1

In a 100 mL round bottom flask containing10 mL of distilled water, was added 2.50 mmol (3.04 mg) of (5-nitro-salicylidene)-2-aminopyridine. This was mixed withcobalt(II) chloride hexahydrate (1.25 mmol, 1.48 mg) in 5 mL distilled water. The combination used in the reaction was agitated at room temperature for one hour, further refluxed for six hours at 60 °C. A brown solid precipitate appeared in the mixture as it cooled to room temperature, and was subsequently collected by filtering. The precipitateproduced 51% (3.92 mg) complex after being cleaned with cold ethanol and dried on silica gel in a desiccator. Melting point: 266 to 269 °C.

IR (cm^{-1}): 3260, 2556, 1687, 1600, 1549, 1501, 1474, 1449, 1346, 1269, 1225, 1179, 1151, 1105, 1021, 981, 846, 824, 745, 703, 679, 648, 512, 471.

Analytical Data: $C_{24}H_{26}CoN_6O_{11}$ was calculated (found) as follows: C, 45.51 (44.98), H, 4.14 (4.09), N, 13.27 (12.67), Co, 9.30 (9.49).

Cobalt(II) Complex of L2

In a 100 mL round bottom flask containing10 mL of distilled water, was added 2.50 mmol (3.45 mg) of (5- bromo-salicylidene)-2-aminopyridine. This was mixed withcobalt(II) chloride hexahydrate (1.25 mmol, 1.48 mg) in 5 mL distilled water. The combination used in the reaction was refluxed for six hours at 60 °C. A brown solid precipitate appeared in the mixture as it cooled to room temperature, and was subsequently collected by filtering. The precipitate was washed 3x with 1:1 (ethanol:water) anddried in a desiccator over silica gel to yield 59% (2.03 mg) product. Melting point: 234 to 237 °C.

IR (cm^{-1}): 3059, 1593, 1566, 1516, 1452, 1373, 1285, 1161, 1100, 979, 927, 872, 831, 805, 772, 743, 698, 565, 472, 467.

Analytical Data: $C_{24}H_{20}Br_2CoN_4O_4$ was calculated (found)as follows: Calculated for C, 44.54 (44.10); H, 3.11 (3.60); N, 8.66 (8.17); Co, 9.11 (9.83).

Larvicidal Activity

Collection and Acclimatization of Test Organisms The primary test organisms were larvae of *Anopheles gambiae* species, including both field-collected and laboratory-bred strains. Field larvae were collected using a 0.05 mm mesh fine sieve net from temporary water pools located on Omotoye Street, Lagos State, Nigeria (geographic coordinates: 6°36'14.5"N, 3°25'45.6"E). The collected larvae were transported to the laboratory for subsequent bioassays. Laboratory-bred *Anopheles gambiae*larvae (KISUMU) were obtained from the Department of Public Health at the Nigerian Institute of Medical Researchand used as collected. Note: The KISUMU strain of *Anopheles gambiae* originates from a village in Kenya, from which it derives its name. KISUMU is not an acronym but a direct reference to the village. This strain is highly susceptible to insecticides and is commonly used as a reference in potency testing of insecticides. When conducting comparative tests between field-collected mosquitoes and the KISUMU strain, it is expected that, under normal conditions, the insecticide should achieve 100% mortality in the KISUMU strain.

Stock Solution Preparation

Stock solutions of the synthesized Schiff base compoundswere prepared by dissolving 0.2 g of the powderedsynthesized compounds in DMSO. The stock solution wassubjected to a range test in order to identify the properdosages or precise dose range. This avoids the use of overlyhazardous ranges that could pose a serious risk to humans. The stock was diluted to give solutions with 250µl/100ml,125µl/100ml, 65.5 µl/100ml, 31.25 µl/100ml, 15.63 µl/100ml, 7.82 µl/100ml, 3.91 µl/100ml, 2 µl/100ml, 1 µl/100ml, 0.5 µl/100ml concentrations. A control experiments were set up using water (positive) and DMSO (negative) controls respectively. Stock solutions were used as freshly prepared stock solution.

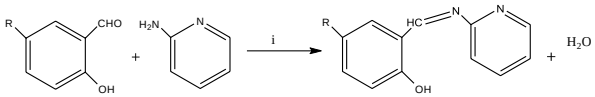
Larvicidal Activity

The susceptibility of *Anopheles gambiae* larvae to the synthesized Schiff base was assessed using a methodology adapted from the Elliot larval test (WHO, 1992). Late thirdand early fourth instar larvae (25 per group) were transferred to small disposable test cups using droppers. A total of 100 mL of various concentrations of the stock solutions was measured with a 100 mL graduated cylinder and added to the cups containing the larvae. Two replicatesof 25 larvae were assigned to each treatment group. A cup containing 100 mL of water served as the positive control, while another cup with 100 mL of dimethyl sulfoxide (DMSO) was used as the negative control. Mortality responses were recorded at 24, 48, and 72 hours. A larva was considered dead if it did not exhibit movement when gently touched with the tip of a pipette or if it failed to rise to the surface or showed no characteristic diving response when the water was disturbed.

Results and Discussion

Synthesis

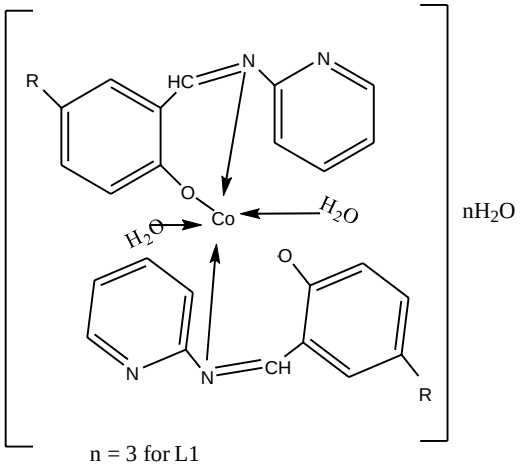
Two Schiff bases— N-(5-nitro-salicylidene)2-aminopyridine **(L1)** and N-(5-bromo-salicylidene)2-aminopyridine **(L2)—**were successfully synthesized via the condensation reaction of 2-aminopyridine with different substituted benzaldehydes. Specifically, **L1** was derived from 5-nitro-salicylaldehyde and **L2** from 5-bromo- salicylaldehyde. Each reaction was carried out under controlled conditions to ensure the efficient formation ofthe Schiff base products, which were then characterized for structural verification through appropriate spectroscopic techniques (Dueke-Eze *et al*., 2011).



Reagents and conditions: (i) Formic acid, ethanol, heat (60-65°C), 6-8 h
R = NO_2 = L1
Br = L2

Scheme 1: Synthesis of 2-aminopyridine based Schiff bases

The reaction of the synthesized Schiff bases with chloride salt of Co(II) produced their corresponding metal complexes (**Figure 1**) in moderate to high yields. These complexes were found to be air-stable and exhibited limited solubility in most common organic solvents, but readily dissolved in polar aprotic solvents such as dimethylformamide (DMF) and dimethyl sulfoxide (DMSO). Comprehensive analytical data for these complexes, summarized in **Table 1**, are in strong agreement with the calculated stoichiometric values, indicating a 1:2 metal-to-ligand ratio.



n = 3 for L1

Figure 1: Proposed structure for the Co(L1) and Co(L2)

Table 1 summarizes the physical and analytical information for the Schiff bases **(L1 and L2)** and their cobalt complexes **(CoL1 and CoL2)**. With minor variations that could be the result of experimental circumstances, the data shows good agreement betweenestimated and experimental values. The stability of the intermediates and the reaction conditions may have contributed to the increased yield of **L2** (81%).

Table 2 presents the IR and NMR results, which shed light on the bonding environment in the Schiff bases and their cobalt complexes. A *ν*C=N stretch at 1595 cm^{-1} is visible in **L1**, indicating the creation of an imine bond. At 1285 cm^{-1}, the *ν*C-O stretch indicates coordination through the oxygen atom. The *ν*C=N and *ν*C-O move to 1600 cm^{-1} and 1269 cm^{-1}, respectively, in **Co(L1),** suggesting coordination to the cobalt center. For **L1**, the NMR data indicates an imine proton shift at 9.53 ppm, accompanied by a matching δC shift at 162.89 ppm. In a similar manner, L2 exhibits *ν*C=N at 1608 cm^{-1} and *ν*C-O at 1276 cm^{-1}, which shifts when **Co(L2)** was coordinated.

There is an imine proton shift for **L1** at 9.53ppm with a δC shift at 160.86 ppm in the NMR data.

Table 3 summarizes the conductivity and electronic spectra, which indicate that the cobalt complexes take on an octahedral shape. Schiff bases exhibit π-π* and n-π* transitions, which are typical of conjugated systems.Charge transfer bands and d-d transitions for **Co(L1)** and **Co(L2)** support the octahedral geometry around the cobalt ion. The low molar conductivity values recorded for all complexes confirm their non-electrolytic nature, suggestingthat they do not dissociate into ions in solution.

Table 1: Physical and analytical data for LI and CoL2

Code	Formula	Mp (°C)	Yeild (%)	Elemental Analysis % Calculated(Found)			
				C	H	N	M
L1	$C_{12}H_9N_3O_3$	182 - 184	46	59.26 (59.14)	3.73 (3. 56)	17.28 (16.96)	-
Co(L1)	$[Co(L1)_2 2H_2O].3H_2O$	266 -269	51	45.51 (44.98)	4.14 (4.09)	13.27 (12.67)	9.30 (9.49)
L2	C_{12} H_9N_2OBr	138-140	81	52.01 (51.96)	3.27 (3.21)	10.10 (9.88)	-
Co(L2)	$[Co(L2)_2.2H2O]$	234-237	59	44.54 (44.10)	3.11 (3.60)	8.66 (8.17)	9.11 (9.83)

Table 2: IR and NMR data for L1 -CoL2

Code	IR bands (cm^{-1})							Chemical shift (ppm) HC=N	
	*ν*OH	*ν*C=N	*ν*C-O	δC=N (py)	*ν*(H_2O)	*ν*(M-O)	*ν*(M-N)	δ_H -	δ_C
L1	3331	1595	1285	1099	-	-	-	9.53	162.89
Co(L1)	-	1600	1269	1105	846	512	471	-	-
L2	-	1608	1276	1100	-	-	-	9.34	160.86
Co(L2)	-	1593	1285	1100	872	472	467	-	-

Table 3: Electronic absorption bands and molar conductance for LI-CoL2

Code	max(nm)				Molar conductance ($ohm^{-1}cm^2mol^{-1}$)	Proposed Geometry
	π-π*	n-π*	CT	d-d		
L1	273	-	373	-	-	-
Co(L1)	-			603, 674	3.63	Octahedral
L2	-	305	358	-	-	-
Co(L2)	-	-		593, 648	3.71	octahedral

Larvicidal Activity

The larvicidal activity of the Schiff bases **(L1 and L2)** and their cobalt complexes **(CoL1 and CoL2)** has been successfully carried out and presented in **Table 4**. Two control experiments were used-water (positive) and DMSO (negative). There was little larval mortality in the water control, indicating low natural mortality in the absence of any chemical. This control further supports the validity of the experiment's design by confirming that handling practices did not significantly contribute to larval death. Larval viability was not adversely affected by DMSO itself, as evidenced by the DMSO solvent control, which displayed zero mortality at all time periods. This further supports the specificity of the larvicide's action by confirming that the mortality seen in treated groups was caused only by the synthesized compounds and not by any control influence. The larvicidal activity test method was a 72-hours toxicity profiling. The level that kills 50% population is considered toxic. A crucial evaluation of the compound's effectiveness across various *Anopheles gambiae* populations was made possible by the inclusion of both KISUMU and Field strains.

The result generally revealed that increased concentrations of the Schiff base compound consistently resulted in increased mortality rates, demonstrating a clear concentration-dependent effect on *Anopheles gambiae*larvae mortality. Total mortality (100%) and 50% mortality was recorded at all time points at the highest tested concentration (250 µl/100 mL) for the metal complexes andthe ligands respectively. This is an indication that this concentrations are extremely harmful to both KISUMU andfield strains. This outcome suggests that the compounds are successful in interfering with vital biological processes required for larval survival, maybe by damaging the structural integrity of larval tissues or interfering with important biochemical pathways.

During the first 24 hours, a high initial mortality was noted at high to moderate values (125 µl/100 mL and 65.5 µl/100 mL). Nevertheless, some larvae survived after the first exposure, indicating a dose-response relationship in which

certain larvae, although in lesser numbers, can survive longer at sublethal concentrations. This demonstrates the compound's effectiveness, but at these concentrations, its effects can gradually decrease as larvae either adapt to lower toxicity levels or are less affected due to metabolic factors. Mortality decreased significantly at lower concentrations (31.25 µl/100 mL and below), with many larvae surviving for up to 72 hours. The decreased mortality at these concentrations might indicate that the compounds are not toxic enough to produce appreciable death below the effective fatal dose. This insight is critical for future applications of these compounds in larval control, as it establishes an effective dosage threshold for achieving desirable mortality levels in *Anopheles gambiae* populations. In comparison of the Schiff bases to their metal complexes, it was observed that the cobalt complex of the bromo- containing Schiff base **(CoL2)** had marginally greater death rates at 31.25 µl/100 mL than nitro containing cobalt complex **(CoL1)** and the ligands **(L1 and L2)** , which were especially apparent after 24 hours. This suggests that **CoL2** may enhance activity at intermediate dosages by enabling a longer-lasting engagement with larval targets. This difference in efficacy at lower doses suggests that cobalt complexation enhances the larvicidal activity, potentially due to increased stability or enhanced uptake of the cobalt-complexed compounds by the larvae (Kumari *et al*., 2021).

The majority of larval deaths happened within the first 24 hours of exposure for the higher concentrations. This suggests that the mortality response was likewise time-dependent and the compounds operate quickly after administration in which the chemical rapidly reduces larval viability. It is noteworthy that the observed persistence of mortality after 24 hours, even at lower doses (31.25 µl/100 mL) in **CoL2** (bromo-containing compound) implies that this compound may have sub lethal effects that delay larval growth by affecting their feeding, motility, or the ability to complete molting cycles (Sharma *et al*., 2016). Such delayed mortality is a favorable characteristic for larvicides, as it means even larvae that initially survive may ultimately succumb before progressing to pupae and adulthood, thus reducing adult mosquito emergence.

Several variables may be responsible for the increased larvicidal activity observed in the Schiff base cobalt complexes. Complexation is known to increase the stability of Schiff bases, making them less vulnerable to hydrolysis or environmental degradation thus improving lipophilicity and bioavailability (Alfonso-Herrera *et al.* 2022). Cobalt complexes may be more lipophilic, which could improve their capacity to enter larval tissue or more efficiently interact with enzymes that are membrane-bound. This enhanced bioavailability may make it possible for **CoL1** and **CoL2** to more effectively reach their molecular targets, which would enhance their larvicidal actions. Because cobalt ions can produce reactive oxygen species (ROS), biological systems may experience oxidative stress (Dayem *et al*., 2017). The larvae may develop ROS as a result of the cobalt complexes **CoL1** and **CoL2**, which could cause cellular damage and eventually death.

Although the exact mode of action of the Schiff base compound on *Anopheles gambiae* larvae is not fully elucidated, Schiff bases are known for their ability to form metal complexes, which may disrupt essential enzymatic processes within the larvae. Schiff base compounds can interfere with key biological molecules, potentially causing oxidative stress or impairing ATP production (Tsacheva *et al.*, 2023), which would quickly incapacitate larvae. Furthermore, Schiff bases may interact with larval nervous systems or cellular membranes, leading to paralysis or structural damage. This interaction could explain the rapid mortality rates observed, especially at higher concentrations, as essential metabolic functions become compromised.

Table 4: No of dead Larvae at various concentration (µl/100ml) post exposure to **L1-CoL2**

Code	Conc. (µl/100ml)	Initial Exposure		24 hours		48 hours		72 hours	
		KISUMU	Field	KISUMU	Field	KISUMU	Field	KISUMU	Field
L1	250	25	25	25	13	25	15	25	17
CoL1		25	25	25	25	25	25	25	25
L2		25	25	25	14	25	16	25	17
CoL2		25	25	25	25	25	25	25	25
L1	125	25	25	25	0	25	6	25	7
CoL1		25	25	25	25	25	20	25	22
L2		25	25	25	0	25	10	25	13
CoL2		25	25	25	25	25	25	25	25
L1	65.5	25	25	17	0	22	3	23	4
CoL1		25	25	20	25	25	17	25	9
L2		25	25	25	0	25	0	25	0
CoL2		25	25	25	17	25	25	25	25
L1	31.25	25	25	6	0	9	2	9	4
CoL1		25	25	9	5	13	7	16	7
L2		25	25	4	4	9	9	11	11
CoL2		25	25	12	13	25	15	25	19
L1	16.63-0.5	25	25	0	0	0	0	0	0
CoL1		25	25	0	0	0	0	0	0
L2		25	25	0	0	0	0	0	0
CoL2		25	25	5	0	0	0	0	0
H_2O	250-0.5	25	25	10-5	10-5	0-0	0-0	0-0	0-0
DMSO	250-0.5	25	25	0-0	0-0	0-0	0-0	0-0	0-0

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Authors' contributions

Cordelia U. Dueke-Eze, Oghenekevwe G. Ebojoh and Tolulope M. Fasina designed, synthesized and characterized the compounds.
Cordelia U. Dueke-Eze and Tolulope M. Fasina interpreted the results obtained from the spectroscopic and analytical techniques.
Cordelia U. Dueke-Eze, David Ajiboye and Kehinde Kemabonta carried the larvicidal assay and interpreted the result.
Cordelia U. Dueke-Eze and David Ajiboye wrote the manuscript.

Conclusion

The synthesis of Schiff bases **(L1-L2)** and their cobalt complexes **(CoL1-CoL2)** has been described. Thepromising larvicidal activity of the Schiff bases and their cobalt complexes against *Anopheles gambiae* has been well highlighted in this work. The findings show both time- and concentration-dependent effects, with considerable lethality maintained across various strains and notably rapid mortality at higher doses. Across concentration levels and exposure durations, both strains showed comparable fatality rates, indicating that the compounds are generally effective against *Anopheles gambiae.* However, there were some minor differences, especially at the concentration of
31.25 µl/100 mL, where the field strain showed slightly greater survival rates after 72 hours than the KISUMUstrain. This variation may be due to genetic traits specific tothe field strain or geographical adaptations that affect how the larvae react to outside substances. Future research on genetic resistance in field populations is also crucial, as environmental exposure or genetic variability could impact susceptibility over time. By highlighting the compound's particular toxicity, the efficient use of controls enhances the validity of these results. These observations justify more research into the compound's mechanism of action and long-term efficacy against potential resistance and offer a strong basis for thinking about its practical implications in malaria vector control.

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