International Journal of Science and Research (IJSR)
ISSN (Online) : 2319-7064
Index Copernicus Value (2015) : 78.96 | Impact Factor (2015) : 6.391

Synthesis of 6-Anilino Derivatives of benzo[a]phenoxazin-5-one and related Aza Analogues via Palladium-catalyzed Buchwald-Hartwig Amination Reaction

Jude Ikechukwu Ayogu[1], Ojarikre Enoo[2], Uchechukwu Chris Okoro[3]

[1]Department of Pure and Industrial Chemistry, Faculty of Physical Sciences, University of Nigeria, Nsukka, Synthetic Organic Chemistry Unit, 41002, Nigeria
Department of Chemistry, University of Canterbury, Christchurch, New Zealand

[2, 3]Department of Pure and Industrial Chemistry, Faculty of Physical Sciences, University of Nigeria, Nsukka, Synthetic Organic Chemistry Unit, 41002, Nigeria

Abstract: ***The synthesis of new 6-anilino derivatives of angular phenoxazine dye was thoroughly investigated. This was accomplished by the condensation of 2-aminophenol with 2,3-dichloro-1,4-naphthoquinone in an alkaline medium to furnish a good yield of the intermediate, 6-chlorobenzo[a]phenoxazine-5-one (6). Buchwald Hartwig cross-coupling of the intermediate with various amines, viz: 2-amino-4-methylpyridine, 2-aminopyrazine, 2-amino-5-nitropyridine, 2-aminopyridazine, 2-amino-3-hydroxypyridine, aniline, 4-nitroaniline, 4-aminophenol and 2-aminophenolvia water-mediated catalyst activation protocol in DMF/toluene solvents mixture at a temperature of 110 ^{o}C yielded the amino derivatives 6 (a-i) respectively. Structures were established by spectral analyses and micro-analytical data.***

Keywords: benzo[a]phenoxazin-5-one, Buchwald-Hartwig amination, condensation, palladium catalysis

1. Introduction

Phenoxazine (**1),** also known as 2,3,5,6-dibenzoxazine is leaf-like crystal which issoluble in ether, benzene, mineral acid and almost insoluble in water. Its solution showspurple or purplish-red fluorescence. It is a three ring structure compound in which twobenzene rings are joined by oxygen and nitrogen atom at non-adjacent positions. The benzophenoxazines have also been prepared and characterized. The reaction of *o*aminophenol with 2,3-dichloro-1,4-naphthoquinonewas pioneered by VanAllen and Reynolds[1].The chemistry of phenoxazine and its derivatives has been extensivelystudied because oftheir versatile utility in industry, agriculture and medicine. In industries, they havebeen applied as organic dyes for dye-sensitized solar cells[2-4], acid-base and redoxindicators, polymerization retardants, metal extractant[5] and chromophoric compound in host guest artificial protonic antenna system[6]. It has been reported that polymers/oligomers containing a phenoxazine unit could be promising conducting andfluorescent materials. [7,8] A report of the applications of the phenoxazines and their angular derivatives cannot be deemed complete without a portray of their medicinalproperties. They are bioactive heterocyclic compounds with spectrum of biological activities ranging from anti-tumour[9-11], anti-proliferative[12-14], anti-viral[15], anti-malaria[16], anti-microbial[17], anti-tubarcular, anti-cataract[5]to anti-bacteria[18]. For example, the phenoxazinederivative, 2-amino-4,4α-dihydro-4α,7-dimethyl-3*H*-phenoxazine-3-one (**2**) prepared by the reaction of 2-amino-5-methylphenol with bovine haemolysates was reported by Shimmamoto et *al*.[11]to inhibit proliferation and induce apoptosis in the human leukemia cell lines K562, HL-60 and HAL-01 in dose-dependent manner. However, Iwala *et al*[15]and Shimizu *et al*.[19] showed that compound (**2)** is handy ininhibition of multiplication of poliovirus in Vero cells between 0.25-2 µg with maximalactivity at 1 µg, and exhibition of antimicrobial activity against non-tubercularmycobacteria respectively. 2-Amino-3*H*-phenoxazin-3-one was reported by Kohno *etal*. [20]to have anti-inflamatory and immunoregulatory properties thereby providing apromising therapeutic strategy for the treatment of T cell-mediated inflammatoryautoimmune disease as well as bacteria-induced-chronic inflammatory disease. Somephenoxazine structural scaffolds are used as valuable pharmacophores for potenttubulin polymerization inhibitors [14]. Naturally occurring phenoxazines are also known. They are found as the central core of a number of natural chemical compounds such as the antibiotic actinomycin D (**2) (Figure 1)** isolated from soil bacteria of the genus *Streptomyces*,[20] the ommochromes like ommatin D (**3)** and xanthomatin (**4)** found in different anthropods and are responsible for coloration in the wings, cuticles and eyes of insects, and theorcein extracted from several species of lichen, commonly known as "orchella weeds", found in various parts of the world. Orcein is a reddish-brown dye, orchil is a purple-blue dye. Orcein is used as a stain in microscopy to visualize chromosomes,[22] elasticfibers, [23]Hepatitis B surface antigens, [24] and copper-associated proteins. [25]

Licensed Under Creative Commons Attribution CC BY

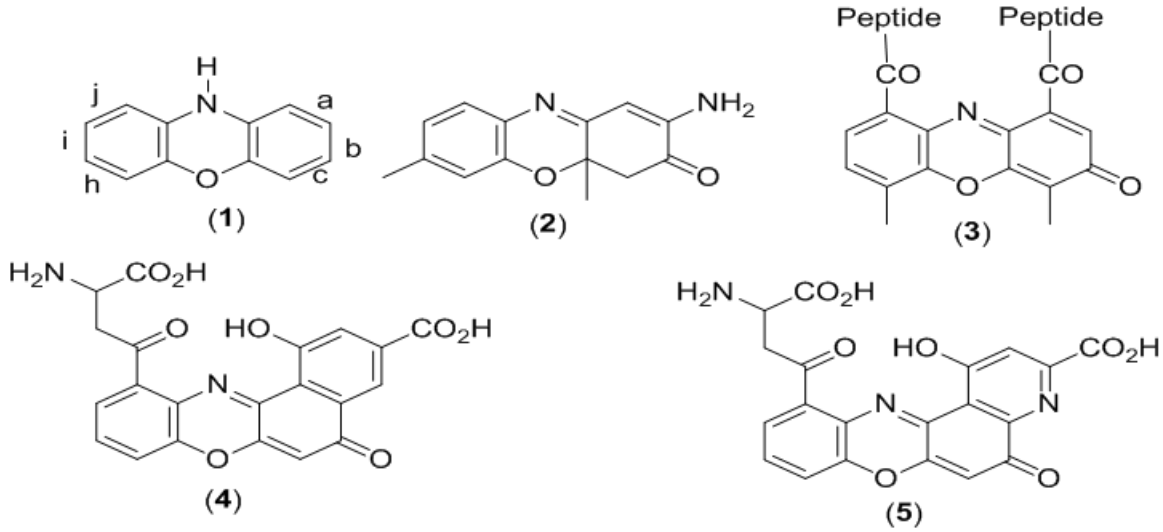


Figure 1

Extensive structural modifications of (**1**) and its derivatives are still in progress in an attempt to obtain new derivatives having more desirable properties. Although several synthetic routes to linear and angular phenoxazines have been reported, methods are often not applicable for the preparation of a wide variety of their derivatives with excellent yields and good pharmacological activity. Moreover, because of the current indispensability of benzophenoxazine rings as valuable molecular motif for the development of chemotherapeutic agents with high pharmacokinetic profile, it becomes imperative to investigate elegant and facile reaction protocols to synthesized possible derivatives with variety of functionalities.

2. Experimental Section

All coupling reactions were carried out under nitrogen atmosphere. Melting points of synthesized compounds were determined with a Fischer John's apparatus and were uncorrected. All compounds were characterized by ^{1}H-NMR, ^{13}C-NMR, IR and UV-Visible spectroscopy. UV-visible spectra were obtained on a UVI ultra-violet spectrophotometer (manufactured by UNICAM, serial number 061408) using marched 1cm quartz cells. The IR spectra were recorded on 8400S Fourier Transform Infra-red spectrophotometer using KBr pellets and the absorption were given in wave numbers (cm^{-1}). The ^{1}H-NMR and ^{13}C-NMR were determined using a Jeol FX 90Q spectrometer using TMS as internal standard (chemical shift in δ). The elemental analysis was carried out with CHN rapid analyzer. The purity of samples was determined using TLC and samples that showed traces of impurities were further purified by column chromatography on silica gel by employing a mixture of ethyl acetate and n-hexane as solvent for elution. Eluates were concentrated using a solvent extractor and air-dried.

6–Chlorobenzo[a]phenoxazin-5-one (6) : To a mixture of 2-aminophenol (4.0 g, 37.00 mmol) and anhydrous sodium acetate (8.2 g, 37.00 mmol) in benzene (100 mL) and DMF (10 mL) warmed to boiling temperature in a 250 mL two-necked flask equipped with magnetic stirrer, thermometer and reflux condenser, was added 2,3-dichloro-1,4-napthoquinone (8.2 g, 37.00 mmol). The entire reaction mixture was refluxed in a water bath at 80 ^{o}C for 6 h with continuous stirring. Thereafter, the reaction mixture was allowed to cool, the content poured into crushed ices (300 g) in a beaker, filtered and air-dried. The crude product was re-crystallized from ethanol-water mixture (2:1) to afford the final product as a yellow solid after *treatment* with activated charcoal. Yield = 6.71 g (65 %) ; m.p: 200-201 ^{o}C (lit. 202-203) [26]; UV-visible (MeOH) λ_{max} (log ε) : 203 (4.33), 209 (4.33), 884 (1.55), 888 (1.50) ; ^{13}C-NMR spectrum (400 MHz, DMSO) δ, ppm: 175.2 (>C=O), 146.7 (Ar-C), 144.7 (Ar-C), 140.7 (Ar-C),138.3 (Ar-C), 138.3 (Ar-C), 136.0 (Ar-C), 132.8 (Ar-C), 130.1 (Ar-C), 118-115 (>C-N-, >C=C<).; ^{1}H-NMR spectrum (400 MHz, DMSO) δ, ppm: 6.70-6.63 (m, 4H, Ar-H), 6.57 (tr, 1H, Ar-H), 6.47 (d, 2 H, Ar-H), 6.45 (tr, 1H, Ar-H) ; IR (KBr) ν: 3371-3254 (N-H), 1650-1573 (>C=O), 1588 (C=C and C=N str) 1371-1288 (C-H bending), 830 (di-substituted benzene), 739 cm-1 (mono-substituted benzene). Anal. cald for $C_{16}H_8ClNO_2$: C 68.16, H 2.84, Cl 12.61, N 4.97, O 11.36; Found: C 68.00, H 2.99, Cl 12.60, N 4.90.

Preparation of the Ligand, 1,4-Bis (2-hydroxy-3,5-di-tert-butylbenzyl) piperazine:A mixture of piperazine (2.2 g, 25.5 mmol) and 40 % aqueous p-formaldehyde solution (4 g, 56.9 mmol) was dissolved in methanol (40 mL) and heated to reflux for 2 h over a water bath and cooled. To the cooled solution was added 2,4-di-tert-butylphenol (10.3 g, 50.41 mmol) in methanol (60 mL). The resulting solution was refluxed for further 12 h at 60 ^{o}C with continuous stirring under a water bath. The reaction mixture was cooled to room temperature and product was re-crystallized from toluene to obtain the pure ligand as a white crystal of melting 260 ^{o}C (lit. >250 ^{o}C) [26]; yield 65%.

Water-mediated Activation of the Palladium Catalyst: To a dried flask flushed with nitrogen gas was added Pd (OAc) $_2$ (0.002 g, 0.001 mmol) dissolved in distilled water (1 mL), 1,4-bis (2-hydroxyl-3,5-di-tertbutylbenzyl) piperazine (0.003 g, 0.003 mmol) and 1,4-dioxane (1 mL). The mixture was heated for 1 min at 80 ^{o}C and then allowed to cool to afford the water promoted active catalyst.

Licensed Under Creative Commons Attribution CC BY

General Procedure for the Buchwald-HartwigCross-coupling: To the substrate 6-chlorobenzo[a]- phenothiazin-5-one (0.282 g, 1.0 mmol), the substituted amine (1.2 mmol) and the base k_2CO_3 (0.194 g, 1.4 mmol) in a mixture of DMF and toluene (2 mL: 2 mL) in a two-necked quick-fit flask equipped with magnetic stirrer, reflux condenser and thermometer, was added the resulting highly active green catalyst solution. The content was refluxed at 110 ^oC with continuous stirring for 3 h on a paraffin oil bath to afford the crude coloured products, **6 (a-i).** Re-crystallization was done from 1:1 heterogeneous mixture of ethyl acetate and water to obtain the pure titled products in various yields. The purity of the product was determined using the thin layer chromatography (TLC) and melting point.

6- (4-Methylpyridin-2-ylamino) benzo[a]pheno-xazin-5one (6a) : As a reddish-brown solid. yield = 0.30 g (85 %) ; m.p: 169-170 ^oC; UV-visible (MeOH) λ_{max} (log ε) : 206 (4.33), 207 (4.41), 879 (1.80), 886 nm (1.78) ; IR (KBr) cm^{-1}: 3533-3300 (NH str), 2980-2850 (CH str), 1635 (>C=O), 1577- 1560 (C=C and C=N str), 1284 (C-O-C), 843 (monosustituted benzene). ^{1}H-NMR (DMSO) δ_H: 7.91 (d, 1H, Ar-H), 7.89 (d, 1H, Ar-H), 7.79 (tr, 1H, Ar-H), 7.77 (d, 1H, Ar-H), 7.67-7.55 (m, 4H, Ar-H), 7.54 (tr, 1H, Ar-H), 6.43 (d, 1H, Ar-H), 5.85 (s, 1H, NH). ^{13}C NMR (DMSO) δ_C: 207.34 (>C=O), 134.19 (Ar-C), 130.95 (Ar-C), 126.0 (Ar-C), 125.78 (Ar-C), 112.30 (>C=N-, >C=C<). Anal. cald for $C_{21}H_{13}N_3O_2$: C 74.26, H 3.84, N 12.38, O 9.42; Found: C 73.99, H 4.00, N 40.75.

6- (Pyridin-2-ylamino) benzo[a]phenoxazin-5-one (6b) : Crystallized as brown solid; yield = 0.25 g (74 %) ; m.p: 160-161^oC; UV-visible (MeOH) λ_{max} (log ε) : 209 (4.41), 207 (4.41), 879 (1.80), 886 (1.78) ; IR (KBr) cm^{-1}: 3533-3300 (NHstr), 2980-2850 (CH str), 1635 (>C=O), 1577-1560 (C=C and C=N str), 1284 (C-O-C), 843 (monosustituted benzene) ;^{1}H-NMR (DMSO) δ_H: 7.91 (d,1H, Ar-H), 7.89 (d, 1H, Ar-H), 7.79 (tr, 1H, Ar-H), 7.77 (d, 1H, Ar-H), 7.67-7.55 (m, 4H, Ar-H), 7.54 (tr, 1H, Ar-H), 6.43 (d, 1H, Ar-H), 5.85 (s, 1H, NH) ; ^{13}C NMR (DMSO) δ_C: 207.34 (>C=O), 134.19 (Ar-C), 130.95 (Ar-C), 126.0 (Ar-C), 125.78 (Ar-C), 112.30 (>C=N-, >C=C<).Anal. cald for $C_{21}H_{13}N_3O_2$: C 74.26, H 3.84, N 12.38, O 9.42; Found: C 73.99, H 4.00, N 40.75.

6- (5-NitroPyridin-2-ylamino) benzo[a]pheno-xazin-5-one (6c) : As reddish-brown solid; yield = 0.17 g (45%) ; m.p: 204-205^oC; UV-visible (MeOH) λ_{max} (log ε) : 205 (4.46), 339 (4.46), 849 (1.58), 886 (1.68) ; IR (KBr) cm^{-1}: 3072-3030 (NH str), 1680-1619 (>C=O), 1534-1440 (C=C and C=N str), 1552-1530 (NO_2), 1264 (C-O-C), 50-600 (monosustituted benzene) ; ^{1}H-NMR (DMSO) δ_H: 8.83-8.82 (d, 3H, Ar-H), 8.10 (d, 1H, Ar-H), 8.09 (tr, 2H, Ar-H), 8.07 (d, 1H, Ar-H), 7.52 (d, 2H, Ar-H), 6.48 (tr, 2H, Ar-H), 3.70 (s, 1H, NH). ^{13}C NMR (DMSO) δ_C: 205.99 (>C=O), 147.86 (Ar-C), 147.53 (Ar-C), 134.9 (Ar-C), 133.10 (Ar-C), 107.70 (>C=N-, >C=C<).Anal. cald for $C_{21}H_{12}N_4O_4$:C 65.57, H 3.13, N 14.57, O 16.65; Found: C 65.50, H 3.19, N 14.50.

6- (Pyrazin-2-ylamino) benzo[a]phenoxazin-5-one (6d) : As an orange brown solid; yield = 0.15 g (45%) ; m.p: 200-201 ^oC; UV-visible (MeOH) λmax (log ε) : 210 (4.41), 206 (4.41), 886 (1.78), 886 (1.78) ; IR (KBr) cm^{-1}: 3440-3280 (NH str), 1680 (>C=O), 1590- 1560 (C=C and C=N str), 1391-1370 (C-H str ofaromatic), 733 (monosustituted benzene) ; ^{1}H-NMR (DMSO) δ_H: 7.90 (d, 1H, Ar-H),7.89 (d, 1H, Ar-H), 7.79 (d, 2H, Ar-H), 7.77 (tr, 1H, Ar-H), 7.67 (d, 2H, Ar-H), 7.54 (tr, 4H, Ar-H), 6.38 (s, 1H, NH), 5.85 (s, 1H, NH). ^{13}C-NMR (DMSO) δ_C: 167.50 (>C=O), 134.20 (Ar-C), 133.02 (Ar-C), 131.21 (Ar-C), 130.95 (Ar-C), 126.01 (Ar-C),125.78 (>C=N-, >C=C<). Anal. cald for $C_{20}H_{12}N_4O_2$: C 70.51, H 3.52, N 16.46, O 9.40;Found: C 70.47, H 3.51, N 16.45.

6- (3-Hydroxypyridin-2-ylamino) benzo[a]- phenoxazin-5-one (6e) : As purple coloured solid; yield = 0.17 g (49%) ; m.p: 190-191 ^oC; UV-visible (MeOH) λmax (log ε) : 224 (4.73), 207 (4.73), 880 (2.06), 888 nm (2.03) ; IR (KBr) cm^{-1}: 3500-3400 (OH), 3349-3261 (NH str), 2940-2920 (C-H str of aliphatic), 1650-1643 (>C=O),1490 (C=C and C=N str), 846-727 cm-1 (monosustituted benzene) ; ^{1}H-NMR (DMSOδ_H:9.50 (d, 1H, Ar-H), 8.99 (d, 1H, Ar-H), 8.49 (tr, 1H, Ar-H), 8.47 (d, 1H, Ar-H), 8.02-7.96 (m, 4H, Ar-H), 7.82-7.74 (tr, 2H, Ar-H), 5.53 (s, 1H, NH) ; ^{13}C NMR (DMSO) δ_C: 180.54-178.09 (>C=O), 158.20 (Ar-C), 157.40 (Ar-C), 135.08 (Ar-C), 133.70 (Ar-C),132.40 (Ar-C), 130.40 (Ar-C), 126.75 (Ar-C), 116.50-116.1 (>C=N-, >C=C<). Anal.cald for $C_{22}H_{13}N_3O_3$: C 70.92, H 3.66, N 11.83, O 13.51; Found: C 71.00, H 3.58, N11.83.

6-Phenylaminobenzo[a]phenoxazin-5-one (6f) : As reddish brown solid; yield = 0.25 g 74%) ; m.p: 134-135 ^oC; UV-visible (MeOH) λmax (log ε) : 224 (3.81), 206 (4.53), 880 (2.04), 891 nm (1.89) ; IR (KBr) cm^{-1}: 3390-3361 (OH), 3349-3261 (NH str), 1645 (>C=O), 1380-1360 (C-H str aromatic), 1590-1560 (C=C and C=N str), 836 (disubstituted benzene) 745 (monosubstitution) ; ^{1}H-NMR (DMSO) δ_H: 7.72 (d, 1H, Ar-H), 7.70 (d, 1H, Ar-H), 7.69 (tr, 1H, Ar-H), 7.51-7.49 (m, 4H, Ar-H), 7.38 (d, 1H, Ar-H), 7.70 (d, 1H, Ar-H), 7.69 (tr, 1H, NH), 7.51-7.49 (m, 4H,Ar-H), 7.38 (d, 1H, Ar-H), 6.84 (m, 4H, Ar-H), 6.30 (s, 1H, NH). ^{13}C-NMR (DMSO) δ_C: 180.80 (>C=O), 149.45 (Ar-C), 148.82 (Ar-C),147.95 (Ar-C),142.49 (Ar-C), 134.30 (Ar-C), 129.39 (Ar-C), 128.50 (Ar-C), 125.85 (Ar-C),116.50-103.99 (>C=N-, >C=C<). Anal. cald for $C_{22}H_{14}N_2O_2$: C 78.03, H 4.14,N 8.28, O 9.46;Found: C 78.03, H 4.14, N 8.29.

6- (4-Nitrophenylamino) benzo[a]phenox-azin-5-one (6 g) : As dark-red solid; yield = 0.32 g (84%) ; m.p: 192-193 ^oC; UV-visible (MeOH) λmax (log ε) : 203 (4.33), 205 (4.53), 879 (1.76), 897 nm (1.89) ; IR (KBr) cm^{-1}: 3484-3348 (NH), 1634-1606 (>C=O), 1560-1471 (C-H str aromatic), 1560 (C=C and C=N), 1471 (NO_2), 1302-1107 (C-H bending), 1293-1014 (C-O-C), 836 (disubstituted benzene) 742 (monosubstitution) ; ^{1}H-NMR (DMSO) δ_H: 7.95 (d, 2H, Ar-H), 7.93 (d, 2H, ArH), 7.69 (d, 2H, Ar-H), 7.53 (tr, 2H, Ar-H), 6.81 (d, 2H, Ar-H), 6.59 (tr, 2H, Ar-H),3.42 (s, 1H, NH) ; ^{13}C NMR (DMSO) δ_C : 156.30 (>C=O), 136.16 (Ar-C), 134.18 (Ar-C), 130.95 (Ar-C), 126.95 (Ar-C), 126.02 (Ar-C), 125.79 (Ar-C), 112.95 (Ar-C),125.85 (>C=N-, >C=C<). Anal. cald for $C_{22}H_{13}N_3O_4$: C 68.87, H 3.39, N 10.96, O16.69; Found: C 69.00, H 3.26, N 10.94.

6- (4-Hydroxyphenylamino) benzo[a]pheno- xazin-5-one (6h) : As dark brown coloured solid;yield = 0.24 g (66%) ; m.p: 192-193 oC; UV-visible (MeOH) λmax (log ε) : 204 (4.33), 207 (3.62), 880 (1.70), 891 nm (1.66) ; IR (KBr) cm^{-1}:3371-3250 (OH), 3436-3254 (NH str.), 1680-1658

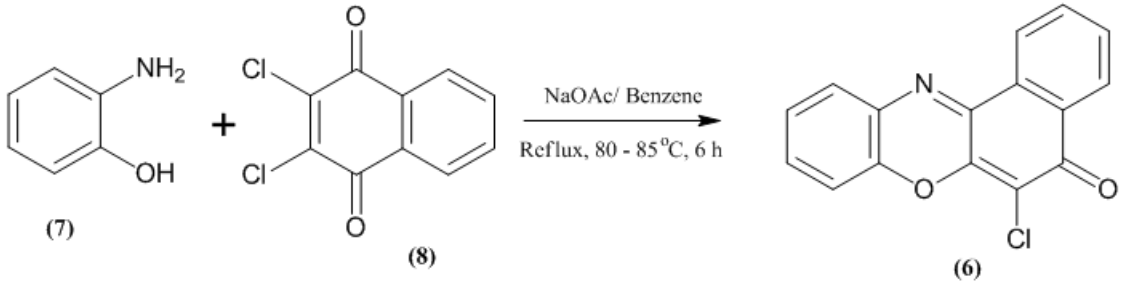
Licensed Under Creative Commons Attribution CC BY

(>C=O), 1590-1560 (C=C and C=N),1371-1288 (C-H bend) 850-830 (*p*-disubstitution), 742 (monosubstitution) ; ^{1}H-NMR (DMSO) δ_H: 8.52 (d, 1H, Ar-H), 8.10 (d, 1H, Ar-H), 7.90 (tr, 2H, Ar-H), 7.8-7.6 (m,4H, Ar-H), 7.50-7.40 (m, 4H, Ar-H), 7.20 (d, 2H, Ar-H), 5.80 (s, 1H, OH) ; ^{13}C NMR (DMSO) δ_C : 134.40 (Ar-C), 130.77 (Ar-C), 125.85 (Ar-C), 125.46 (Ar-C). Anal. caldfor $C_{22}H_{14}N_2O_3$: C 74.50, H 3.95, N 7.91, O 13.55; Found: C 73.99, H 3.97, N 7.95.

6- (2-Hydroxyphenylamino) benzo[a]-phenoxazin-5-one (6i) : As black coloured solid; yield = 0.30 g (84%) ; m.p: 204-205^{o}C; UV-visible (MeOH) λ_{max} (log ε) : 203 (log ε 3.42), 220 (log ε 4.53), 877 (log ε 1.79), 897 nm (log ε 1.68). IR (KBr) cm^{-1}: 3440-3432 (NH), 2940-2920 (C-H str.), 1681-1672 (>C=O), 1590-1560 (C=C and C=N), 742 (monosubstitution) ; ^{1}H-NMR (DMSO) δ_H: 7.90 (d, 1H, Ar-H), 7.89 (d, 1H, Ar-H), 7.79 (tr, 2H, Ar-H), 7.77 (d, 2H, Ar-H), 7.67 (tr, 2H, Ar-H), 7.55 (d, 2H, Ar-H), 7.53 (Ar-C), 6.3 (s, 1H, OH). ^{13}C-NMR (DMSO) δ_C: 167.60 (>C=O), 135.71 (Ar-C), 134.15 (Ar-C), 130.93 (Ar-C), 125.99 (Ar-C), 125.77 (Ar-C).Anal. cald for $C_{22}H_{14}N_2O_3$: C 74.50, H 3.95, N 7.91, O 13.55; Found: C 73.99, H 3.97, N 7.95.

3. Result and Discussion

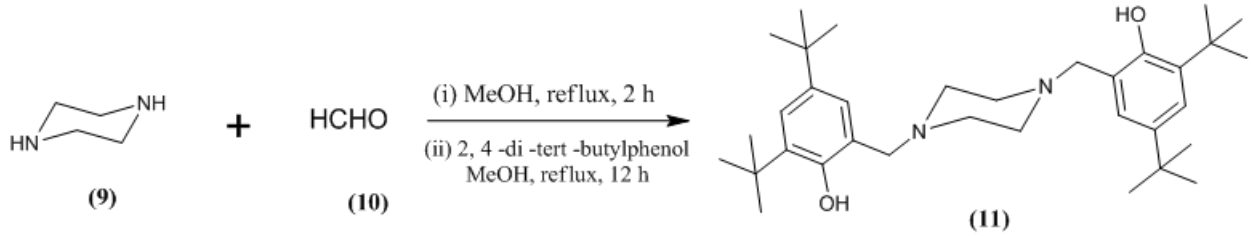
The intermediate, **6** was prepared by the condensation of 2-aminophenol (**7**) with 2,3-dichloronaphthalene-1,4-dione (**8)** in a basic medium using benzene/ DMF as the solvent (Scheme **1**). The product was obtained as shiny yellowish solid after recrystallization from water-methanol. The structure assigned to the intermediate was consistent with their spectra and these were confirmed by the microanalysis.



(7) (8) (6)

Scheme 1

The coupling of the intermediate with the various substituted amines was achieved by the initial activation of the catalyst. A highly active catalyst was generated by heating the Pd (OAc) $_2$in water with 1,4-bis (2-hydroxy-3,5-di-tert-butybenzyl) - piperazine ligand (**11)** for 60 s at 80 ^{o}C in 1,4-dioxane. The ligand **11** was first generated by refluxing a mixture of piperazine**9** and *p*-formaldehyde **10 (**40 % solution) dissolved in methanol for 2 h over a water bath followed by the addition of methanolic solution of 2,4-di-tertbutyphenol (Scheme **2**) as described in the experimentation section. The pre-activation was monitored by colour change from initial yellow to red, dark-red and then to greenish black in 0, 15, 30 and 60 min respectively. Consequently, the palladium (II) was reduced to the active palladium (0) in situ.



(9) (10) (11)

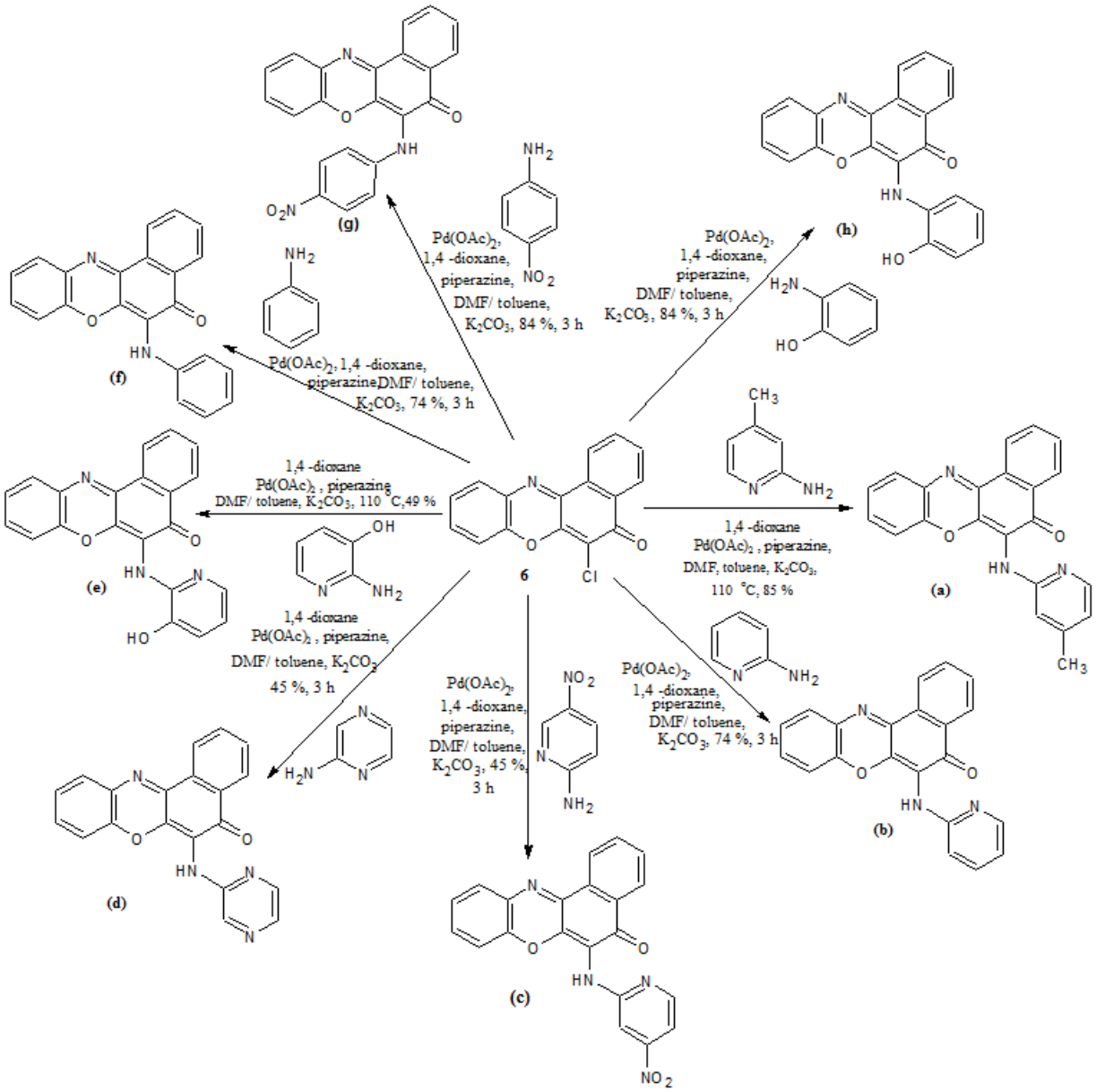
Scheme 2

Thereafter, the active palladium catalyst was applied in the successful synthesis of benzo[a]phenoxazine derivatives (**6a-i).** The catalytic system in combination with either sodium acetate or K_3CO_3 and non-polar solvents (toluene) with little DMF (5mL) was employed in the preparation of the derivatives. The preliminary experiment was started by coupling 6-chloro-benzo[a]phenothiazine 285 mg (1 mmol) with 2-amino-4-methylpyridine 1.2 (mmol) and K_3CO_3 (3 mmol), and the reaction was run intoluene at 110 ^{o}C while the progress of reaction was monitored on hourly basis by TLC. Product formation was observed after 1 h with trace conversion which increased with time. However, after 3 h there was no change in TLC spot of product even after the reaction was left to run overnight. Isolated product yield of 85% with a conversion of over 80% was obtained after work-up and purification by column chromatographyon silica gel using EtOAc/water solvent mixture (1:1). The spectra and elemental analysis confirmed the identity of the isolated product of the coupling reaction as 6- (4-methylpyridin-2-ylamino) benzo[a]- phenoxazin-5-one (**6a)** with molecular formula$C_{22}H_{15}N_3O_2$.

The structure assigned to the 6- (4-methylpyridin-2-ylamino) benzo[a]pheno-xazin-5-onewas consistent with the UV-visible, IR, ^{1}H-NMR and ^{13}C-NMR spectra. The structural assignment was confirmed by the micro-analytical result of compound. The reaction ran without addition of the ligand practically gave no conversion even when it was left over night (24 h). Consequently, the ligand plays a crucial role in Buchwald-Hartwig cross-coupling reactions. While the use

Licensed Under Creative Commons Attribution CC BY

of Na_2CO_3 gave lower product yields (24%), K_3PO_4 gave comparable yield to K_2CO_3 under the same conditions. It was also noticed that the reactions performed without prior solvent degassing with nitrogen gave comparable yield of products under these conditions. Although only Pd $(OAc)_2$ was used as the palladium source; it was believed that other palladium species such as PdCl2, Pd $(NO)_2$ and Pd_2 $(dba)_3$ together with the ligand would make similar transformations. Consequently, the procedure was applied in coupling other intermediate to the other amines running the reaction for a maximum of 3 h under reflux (**scheme 3**) Each completed reaction was work-up by first extracting the crude product from water with dichloromethane followed by purification by column chromatography on silica gel. The molecular structures of the synthesized compounds were established on the basis of IR, NMR, UV and elemental analysis which were in agreement with the assigned molecular structures and formula. The numbers of aromatic protons were accounted for in the 1H-NMRof the synthesized compounds and were found at 9.50-6.43 ppm. The carbon nuclei were nicely represented in the carbon nuclear magnetic spectra with the carbonyl carbon signals found in a far distance low field from the others sp2 hybridized carbon peaks except in **6 d, 6 g** and **6h**. The IR absorption maxima for carbonyl functional groups in synthesized compounds were in the range of 1681-1606 cm^1.Furthermore, the UV-visible spectra data of compounds revealed bathochromic shifts as a result of increase in conjugation with compound **6 g** exhibiting the highest shifts. However, results from elemental analysis of compounds undoubtedly confirmed their molecular and structural formulae assignment.



Scheme 3

Licensed Under Creative Commons Attribution CC BY

4. Conclusion

The present study has shown that the palladium-catalyzed Buchwald-Hartwig cross-coupling reaction offers an excellent route in the preparation of 6-anilino derivatives of angular phenoxazine dyes, **6 (a-h).** Hence, the study has opened the window for further research to ascertain the medicinal potentials of these compounds as well as their applications in solar cells as sensitizers.

References

[1] Van Allen, J. A.; Reynolds, G. A. Polynuclear Heterocycles. VII. The Properties of Azacarbons Containing the Enol Betaine Structure. *J. Org. Chem.*1963, 28, 1022-1025.

[2] Li, J., Yang, X., Cheng, M., Wang, M., Sun, L. Phenoxazine-based Panchromatic Organic Sensitizers for Dye-sensitized Solar Cells. *Dyes and Pigments* 2015, 116, 58-64.

[3] Karlsson, M.; Yang, L.; Karlsson, M. K.; Sun, L.;Boschloo, G.; Hagfeldt, A. Phenoxazine Dyes in Solid-state Dye-sensitized Solar Cells. *Journal of Photochemistry and Photobiology A: Chemistry* 2012, 239, 55–59.

[4] Anil Reddy, M.; Vinayak, B.; Suresh, T.; Niveditha, S.; Bhanuprakash, K.; Prakash Singh, S.; Islam, A.; Han, L.; Chandrasekharam, M. Highly Conjugated Electron rich Thiophene Antennas on Phenothiazine and Phenoxazine-based Sensitizers for Dye Sensitized Solar Cells. *Synthetic Metals* 2014, 195, 208.

[5] Okafor, C. O. Synthesis, Properties and Uses of Angular Phenoxazines. *Dyes andPigments*1986, 7, 103-131.

[6] Mickevičienė, K.; Baranauskaitė, R.; Kantminienė, K.; Stasevych, M.; Komarovska Porokhnyavets, O.; Novikov, V. Synthesis and Antimicrobial Activity of *N* Substituted-β-amino Acid Derivatives Containing 2-hydroxyphenyl Benzo[b]phenoxazine and Quinoxaline Moieties *Molecules* 2015, 20, 3170.

[7] Yuanfu, P.; Otake, M.; Vacha, M.; Sato, H. Synthesis and Characterization of a Novel Electroluminescent Polymer Based on Phenoxazine and Fluorene Derivatives.
React. Funct. Polym. 2007, 67, 1211–1217.

[8] Idzik, K.; Sołoducho, J.; Łapkowski, M.; Golba, S. Development of structural Characterization and Physicochemical Behaviour of Triphenylamine Blocks. *Electrochimica Acta* 2008, 53, 5665-5669.

[9] Koshibu-Koizumi, J.; Akazawa, M.; Iwamoto, T.; Takasaki, M.; Mizuno, F.; Kobayashi, R.; Abe, A.; Tomoda, A.; Hamatake, M.; Ishida, R. Anti-tumour Activity of a Phenoxazine Compound, 2-amino-4,4α-dihydro-4α,7-methyl-3*H*phenoxazine-3-one against Human B cell and T cell Lymphoblastiod Cell Lines: Induction of Mixed Types of Cell Death, Apoptosis, and Necrosis. *J. Cancer Rev. Clin. Onclo*. 2002, 128, 363.

[10] Motohashi, N.; Mitscher, L. A.; Meyer, R. Potential Anti- tumour Phenoxazines. *Med. Res. Rev*. 1991, 11, 239-294.

[11] Shimamoto, T.; Yamaguchi, T.; Sato, K.; Izumi, R.; Tomoda, A. Anti-tumour Effects of a Novel Phenoxazine Derivative on Human Leukemia Cell Lines in vitro and in vivo. *Clin. Cancer Rev.* 2001, 704, 704-708.

[12] Ike, C.O., Ezema, B.E.; Ayogu, J. I.; Ugwu, D.I.; Ezema, C.G.Nwasi, A.P.; Adekola, E. O. Synthesis and Antimicrobial Screening of Novel Benzoxazinophenothiazine Derivatives. *Asian Journal of Chemistry* 2015, 27, 4115-4119

[13] Horton, J. K.; Thimmaiah, K. N.; Harwood, F.C.; Kuttesch, J. F.; Houghton, P. J.Pharmacological Characterization of *N*-substituted Phenoxazines Directed towardReversing Vinca Alkaloid Resistance in Multidrug-resistant Cancer Cells. *Mol. Pharmacol.* 1993, 44, 552-559.

[14] Prinz, H.; Chamasmani, B.; Vogel, K.; Böhm, K. J.; Aicher, B.; Gerlach, M.; Günther, E. G.; Amon, P.;, Ivanov, I.; Müller, K. *N*-benzoylated Phenoxazines and Phenothiazines: Synthesis, Anti-proliferative Activity, and Inhibition of Tubulin Polymerization, *Journal of Medicinal Chemistry* 2011, 54, 4247.

[15] Iwata, A.; Yamagushi, I.; Sato, K.; Izumi, R.; Tomoda, A. Antiviral Activity of 2-Amino-4,4-α-dihydro-4α,7-methyl-3*H*-phenoxazine-3-one on Poliovirus. *Yohoku J. Exp. Med.* 2013, 200, 161.

[16] Mizukawa, Y.; Ge, J.-F.; Bakar M, A.; Itoh, I.; Scheurer, C.; Wittlin, S.; Brun, R.;Matsuoka, H.; Ihara, M. Novel Synthetic Route for Antimalarial Benzo[a]phenoxazine Derivative SSJ-183 and Two Active Metabolites. *Bioorganic and Medicinal Chemistry* 2014, 22, 3749.

[17] Mickevičienė, K.; Baranauskaitė, R.; Kantminienė, K.; Stasevych, M.; Komarovska Porokhnyavets, O.; Novikov, V. Synthesis and Antimicrobial Activity of *N*-substituted-β-amino Acid Derivatives containing 2-Hydroxyphenyl benzo[b]phenoxazine and Quinoxaline Moieties. *Molecules* 2015, 20, 3170.

[18] Ayogu,J. I.; Ezema, B. E.; Ezema, C. G. Utility of Suzuki-Miyaura Cross-Coupling Reaction in the Synthesis of Benzo[a]phenothiazine and Benzo[a]phenoxazine Derivatives and their Anti-microbial Screening. *Asian Journal of Chemistry* 2015, 27, 4467-4473.

[19] Shimizu, S.; Suzuki, M.; Tomoda, A.; Arai, S.;Taguchi, H.; Hanawa, T.; Kamiya, S. Phenoxazine Compounds Produced by the Reactions with Bovine Hemoglobin Show Antimicrobial Activity against Non-tuberculosis Mycobacteria. *Tohoku J. Exp. Med.* 2004, 203, 47-52.

[20] Kohno, K.; Miyake, M.; Sano, O.; Tanaka-Kataoka, M.; Yamamoto, S.; Koya-Miyata, S.; Arai, N.; Fujii, M.; Watanabe, H.; Ushio, S.; Iwaki, K.; Fukuda, S. Anti-inflammatory and Immunomodulatory Properties of 2-Amino-3*H*-phenoxazin-3-one. *Biol. Pharm. Bull.* 2008, 31, 1938-1945.

[21] Hollstein, U. Actinomycin Chemistry and Mechanism of Action. *Chemical Reviews* 1974, 74, 625–652.

[22] La Cour, L. Acetic-orcein: a New Stain-fixative for chromosomes. *Stain Technology* 1941, 16, 169–174.

[23] Friedberg, S. H.; Goldstein, D. J. Thermodynamics of orcein staining of elastic fibres. *Histochemical Journal* 1969, 1, 261–376.

[24] Fredenburgh, J. L.; Edgerton, S. M.; Parker, A. E. 1978. A Modification of the Aldehyde Fuchsin and Orcein Stains for Hepatitis B Surface Antigen in Tissue and a Proposed Chemical Mechanism. *Journal of Histotechnology* 1978, 1, 223–228.

Licensed Under Creative Commons Attribution CC BY

[25] Henwood, A. Current Applications of Orcein in Histochemistry. A Brief Review with some New Observations Concerning Influence of Dye Batch Variation and Aging of Dye Solutions on Staining. *Biotechnic & Histochemistry* 2003, 78, 303–308.
[26] Okafor, C.O. Studies in the Heterocyclic Series XII. The Chemistry and Applications of Aza- and Thia-analogues of Phenoxazine and Related Compounds, *Heterocycles* 1977, 7, 1192.
[27] Mohanty, S.; Sureh, D.; Balakrishna, M. S.; Mague, J. T. An Inexpensive and Highly Stable Ligand, 1,4-bis (2-hydroxy-3,5-di-tert-butylbenzyl)piperazine for Mizoroki-Heck and Room Temperature Suzuki-Miyaura Cross-coupling Reactions. *Tetrahedron* 2008, 64, 240-247.

Licensed Under Creative Commons Attribution CC BY