Research Article

Synthesis of some chloro-substituted isoxazoline derivatives as antibacterial agents

Nagesh J. Deshmukh, Gajanan D. Kottapalle, Avinash T. Shinde*

PG research center department of Chemistry, N.E.S. Science College, Nanded. (Maharashtra)- 431602 (India)

Received: 17 August 2018 Revised: 5 September 2018 Accepted: 24 September 2018

Abstract

Objectives: The chemistry of chalcones has generated intensive scientific studies throughout the world. Especially interest has been focused on the synthesis and biodynamic activities of isoxazoline derivatives. In recent year numerous isoxazoline derivatives have been synthesized for their anticancer, anti-inflammatory, cytotoxic, antiviral activities. The reaction of substituted chalcones with hydroxylamine hydrochloride in presence of acetic acid gave isoxazoline derivatives. **Material and methods**: The five membered oxygen and Nitrogen containing compounds exhibited antioxidant, analgesic, antibacterial, antifungal activities. Therefore the attempt have been made to synthesize chloro-substituted isoxazolines by the reaction of different substituted chalcones with hydroxylamine hydrochloride in presence of few drops of acetic acid. All the synthesized compounds confirmed by TLC and spectral analysis and also screened for their antibacterial activity. **Results**: The novel chloro-substituted isoxazolines showed good to moderate antibacterial activity against Gram+ve and Gram –ve bacterial strains tested. **Conclusion**: These compounds containing chloro, bromo, iodo groups were found showed potent antibacterial, antifungal activities. These findings promoted us to to synthesize novel chloro-substituted isoxazolines derivatives which are found to be potential antibacterial agents.

Keywords: Chalcones, hydroxylamine hydrochloride, isoxazolines, antibacterial activity

Introduction

Heterocyclic compounds are widely distributed in nature and are essential to life in various ways, particularly these compounds are important because of the wide variety of physiological activities associated with this class of substances. Heterocyclic rings are present in several compounds. Most of the members of vitamins B-complex, antibiotics, chlorophyll, haemin, amino acid, enzymes, plant pigments, dye stuff, genetic material DNA etc.

The glorious importance of heterocycles in natural product chemistry and pharmacology constantly drive the search for new methods for the construction of heterocyclic molecules containing azole unit such as pyrazoles and isoxazoles. These pyrazole and isoxazoles were prepared from chalcones which are important intermediate products and they also possess biological and pharmacological applications (Dhar, 1981). The substituted azole unit is an essential pharmacophore of number of antifungal (Chevreuil et al., 2007), antibacterial (Solanki and wadodkar, 2003) and various biological activities(Gautam, 2013; Patel, 2017; Dongre, 2017; Ali, 2011) Therefore considering the antifungal and antibacterial potential of azole derivatives (Sharma and Sharma, 2010), we would like to report herein the synthesis of new isoxazolines and evaluation of their antibacterial activity against *E- coli,S. typhi* (Gram–ve) and *P. aeruginosa, S. aureus* (Gram +ve) bacterial strain by using disc diffusion method (Shinde et al., 2016).

Material and methods

All solvents and chemicals were purchased from Alfa acer chemicals and used without further purification. Melting points were determined in an open capillary tube and are uncorrected. IR spectra were recorded in KBr on a Perkin-Elmer spectrometer. ^{1}H NMR spectra were recorded on a Gemini 300-MHz instrument in $CDCl_3$ as solvent and TMS as an internal standard. The mass spectra were recorded on EISHIMADZU-GC-MS spectrometer. Elemental analysis was carried out on a Carlo Erba 1108 analyzer. The purity of products was checked by Thin Layer Chromatography (TLC) on silica gel.

Address for Corresponding Author:
Avinash T. Shinde
PG Research Center Department of Chemistry, N.E.S. Science College, Nanded. (Maharashtra)- 431602 (India)
E-mail address: drats04@gmail.com

DOI: https://doi.org/10.31024/ajpp.2019.5.1.6

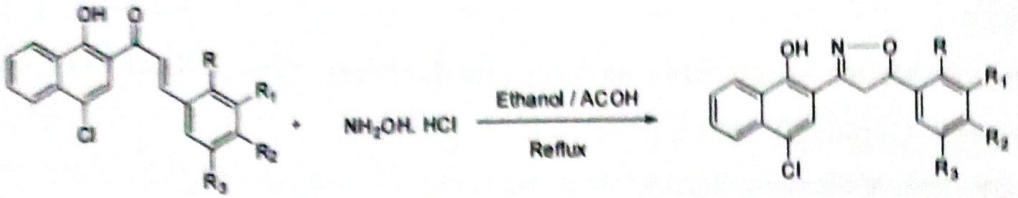

Figure 1. Synthesis of Isoxazolines

5a. **R**= H, R_1 = OCH_2CH_3, R_2= OH, R_3=H

5b. **R**= H, R_1=OCH_2CH_3, R_2= OH, R_3=Br

5c. **R**= O-CH_3, R_1=H, R_2= H, R_3=Cl

5d. **R**= OH, R_1=H, R_2= Cl, R_3=H

5e. **R**= OH, R_1= R_3= Br, R_2=H

5f. **R**= OH, R_1= R_3= I, R_2=H

General procedure for synthesis of Isoxazolines

A mixture of substituted chalcones (0.01mol) and hydroxyamine hydrochloride (0.02 mol) in 20 ml ethanol was refluxed for 5-6 hr in presence of 2-3 drops of acetic acid. After completion of the reaction the reaction mixture was cooled and poured into ice cold water. The resultant solid product (5a-5f) was filtered, washed with sufficient cold water, dried and purified by recrystallization from ethanol.

Results and discussion

All the synthesized compounds (**5a-5f**) have been characterized by their M.P., Elemental analysis, IR, ^{1}H NMR and mass spectra.

4-chloro-2-(5-(3-ethoxy-4-hydroxyphenyl)-4,5-dihydroisoxazol-3yl) naphthalene-1-ol. (5a):

M.F: $C_{21}H_{18}ClNO_4$, M. Wt : 383.82, M.P : 190^{0C}, Elemental analysis (%): C- 65.71, H- 4.73, N- 3.65, Cl- 9.24, IR (Cm^{-1}): 3414(OH), 1498 (C=N), 1382 (C-O), ^{1}HNMR (300 MHz $CDCl_3$) ppm: 1.32 (t,3H, CH_3), 4.09(q, 2H, O-CH_2), 3.60(dd, 1H, CH_A), 3.85(dd, 1H, CH_B), 5.93(dd, 1H, CH_X), 6.75-8.63(m, 8H, Ar-H), 13.10(s, 2H, OH).

2- (5-(3-bromo-5-ethoxy-4hydroxyphenyl)4,5-dihydroisoxazol-3yl)-4-chloronaphthalen-1-ol. (5b):

M.F: $C_{21}H_{17}BrClNO_4$, M. Wt : 462.72, M.P : 195^{0C}, Elemental analysis (%): C- 54.51, H- 3.70, N- 3.03, Cl- 7.76, Br- 17.27, IR (Cm^{-1}): 3473(OH), 1506 (C=N), 1380 (C-O), ^{1}HNMR (300 MHz $CDCl_3$) ppm: 1.33 (t,3H, CH_3), 4.12(q, 2H, O-CH_2), 3.61(dd, 1H, CH_A), 3.88(dd, 1H, CH_B), 5.93(dd, 1H, CH_X), 6.90-8.63(m, 7H, Ar-H), 13.10(s, 2H, OH).

4-chloro-2-(5-(5-chloro-2methoxyphenyl) -4,5-dihydroisoxazol-3yl) naphthalene-1-ol. (5c):

M.F: $C_{20}H_{15}Cl_2NO_3$, M.Wt : 388, M.P : 191^{0C}, Elemental analysis (%): C- 61.87, H- 3.89, N- 3.61, Cl- 18.26, IR (Cm^{-1}): 3380(OH), 1510 (C=N), 1385 (C-O), ^{1}HNMR (300 MHz $CDCl_3$) ppm: 3.83 (s,3H, OCH_3), 3.60(dd, 1H, CH_A), 3.84(dd, 1H, CH_B), 5.94(dd, 1H, CH_X), 6.86-8.63(m, 8H, Ar-H), 12.10(s, 1H, OH).

4-chloro-2-(5-(5-chloro-2-hydroxyphenyl) -4,5-dihydroisoxazol-3yl) naphthalene-1-ol. (5d):

M.F: $C_{19}H_{13}Cl_2NO_3$, M.Wt : 374.22, M.P : 188^{0C}, Elemental analysis (%): C- 60.98, H- 3.50, N- 3.74, Cl- 18.95, IR (Cm^{-1}): 3421(OH), 1510 (C=N), 1375 (C-O), ^{1}HNMR (300 MHz $CDCl_3$) ppm: 3.62(dd, 1H, CH_A), 3.86(dd, 1H, CH_B), 5.95(dd, 1H, CH_X), 6.82-8.63(m, 8H, Ar-H), 12.90(s, 1H, OH).

4-chloro-2-(5-(3,5-dibromo-2-hydroxyphenyl) -4,5-dihydroisoxazol-3yl) naphthalene-1-ol. (5e):

M.F: $C_{19}H_{12}Br_2ClNO_3$, M.Wt : 497.56, M.P : 201^{0C}, Elemental analysis (%): C- 45.86, H- 2.43, N- 2.82, Cl- 7.13,Br- 32.12, IR (Cm^{-1}): 3435(OH), 1498 (C=N), 1370 (C-O), ^{1}HNMR (300 MHz $CDCl_3$) ppm: 3.61(dd, 1H, CH_A), 3.84(dd, 1H, CH_B), 5.96(dd, 1H, CH_X), 7.02-8.64(m, 7H, Ar-H), 12.96(s, 2H, OH).

4-chloro-2-(5-(2-hydroxy-3,5-diiodophenyl) -4,5-dihydroisoxazol-3yl) naphthalene-1-ol. (5f):

M.F: $C_{19}H_{12}I_2ClNO_3$, M.Wt : 591.57, M.P : 211^{0C}, Elemental analysis (%): C- 38.58, H- 2.02, N- 2.36, Cl- 5.99, I- 42.90, IR (Cm^{-1}): 3414(OH), 1508 (C=N), 1375 (C-O), ^{1}HNMR (300 MHz $CDCl_3$) ppm: 3.60(dd, 1H, CH_A), 3.84(dd, 1H, CH_B), 5.96(dd, 1H, CH_X), 6.98-8.64(m, 7H, Ar-H), 12.96(s, 2H, OH.

Antibacterial activity

All the newly synthesized compounds were screened in vitro antibacterial activity. The antibacterial activity was evaluated against 24hr culture of different bacterial strains such as *E- coli, S. typhi* (Gram –ve) and *P. aeruginosa, S. aureus* (Gram +ve) at a concentration 50 µg ml^{-1}. The cultures were diluted with 5% of autoclaved saline and the final volume was adjusted to a concentration of approximately 105-106 CFU ml-1. The synthesized compounds were diluted with acetone for the antibacterial biological assay for agar disc diffusion method. The liquid form of test compound was soaked on to a disc (5mm) and then allowed to air dry, such that the disc became completely saturated with the test compound. The saturated

Table 1. Antibacterial activity (zone of inhibition mm) of compounds (**5a-5f**)

Compounds	Diameter of zone of inhibition (mm)			
	Gram +ive bacteria		Gram -ive bacteria	
	P. aeruginosa	*S. aureus*	*S. typhi*	*E. coli*
5a	23	21	24	24
5b	25	22	23	26
5c	26	24	22	28
5d	25	23	26	27
5e	19	15	20	22
5f	19	14	16	22
Standard	27	26	28	30
DMSO	-	-	-	-

Positive control (Standard): Ofloxacin; Negetive control: DMSO

chemical discs were introduced onto the upper layer of medium evenly loaded with the bacteria and incubated at 37^{0}C for 24 to 48 hrs for better inhibition of bacteria. The zones of inhibition were measured after 24 to 48 hrs. All the experiments were performed in triplicate and the results are expressed as zone of inhibition in mm. The zone of inhibition of the synthesized compounds (**5a-f**) was compared with zone of inhibition of standard antibiotics Ofloxacin (50 µg mL-1).

From the screening studies (**Table 1**), it is evident that the synthesized isoxazoline derivatives **5a, 5b, 5c** and **5d** showed good antibacterial activity against all the tested organisms. It was further observed that the electron rich (**5c**) with one –OMe and two -Cl substituent, showed best activity near to that of standard drug. This observation leads to conclusion that electron rich isoxazolines showed higher activity against bacterial strain tested.

Conclusion

In the present work, we synthesized some novel isoxazoline derivatives from different substituted chalcones and hydroxylamine hydrochloride. The newly synthesized compounds were obtained in good yield and confirmed by spectral analysis. Ofloxacin is used as standard drug for antibacterial activity. The antibacterial data revealed that all compounds showed good to moderate activity compared to standard drug.

Conflict of interest

There is no conflict of interest in the present study.

References

Ali MA, Ismail R, Choon TS, Pandian S, Ansari MZH. 2011. Antimycobacterial activity: Synthesis of novel 3-(substituted phenyl)-6,7- dimethoxy-3a,4-dihydro-3H- indeno(1,2-c) isoxazole analogues. Journal of Enzyme Inhibition and Medicinal Chemistry, 26(4): 598-602.

Chevreuil F, Landreau A, Seraphin D, Larcher G, Mallet S, Bouchara J, Richomme P. 2007. Synthesis of new isoxazoles and dihydroisoxazoles and in vitro evaluation of their antifungal activity. Journal of Enzyme Inhibition and Medicinal Chemistry, 22(5):563-569.

Dhar DN. 1981. The Chemistry of Chalcones and related compounds, John Wiley & Sons Inc.

Dongre RP, Rathod SD. 2017. Synthesis of novel isoxazoline derivatives containing s-Triazine via chalcones and their antimicrobial studies. Der Pharma Chemica, 9(7):68-71.

Gautam KC, Singh DP. 2013. Synthesis and antimicrobial activity of some isoxazole derivatives of thiophene. Chemical Science Transactions, 2(3):992-996.

Patel D, Kumari P, Patel NB. 2017. Synthesis and biological evaluation of coumarin based isoxazoles, pyrimidines and pyrimidin-2-ones. Arabian Journal of Chemistry, 10:3990-4000.

Sharma V, Sharma KV. 2010. Synthesis and biological activity of some 3,5-diarylisoxazolines derivatives: Reaction of substituted chalcones with hydroxylamine hydrochloride. E-Journal of Chemistry, 7(1):203-209.

Shinde AT, Deshmukh NJ, Kottapalle GD, angade SBZ. 2016. synthesis and antimicrobial evaluation of some new fluoro-formazans. Journal of Pure and Applied Chemistry Research, 5(2):61-66.