Synthesis, Antibacterial and Antifungal activity of some Novel Oxadiazole-Pyrazolyl derivatives

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Abstract

Ten title compounds(2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl)-N-(((S)-1-(4-acetyl-4,5-dihydro-5-methyl-5-phenyl-1,3,4-oxadiazolyl)ethylcarbamoyl)methyl)]acetamide,1a-1J, were synthe sized from the starting material (2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl)acetamido ethanamido] propyl hydrazide by treatment with substituted ketones . The synthesized compounds were characterized by physical constants, and the structures of the title compounds were further confirmed by IR, 1H NMR, 13C NMR and elemental analysis. The bioassay results showed that title compounds possessed weak to good anti-microbial activity with 1e showing significant enhancement of bacterial resistance.
Key wards: hydrazides, Ketones, antimicrobial Elemental analysis.

Introduction

One of the major objectives of organic and medicinal chemistry is the design, synthesis and production of molecules, which are having highly therapeutic interest. On the other hand because of the resistance of pathogenic bacteria towards available anti- biotics is rapidly becoming a worldwide indigestive problem, in the same way fungal infections continue to increase rapidly because of the increased number of immune compromised patients. So in view of the above discussions it will be necessary to design a new class of molecules to deal with resistant bacteria and fungi has become one of the most important areas of antimicrobial research today. Since the discovery of heterocyclic nucleus the chemistry of isoxazole and their fused derivatives continue to draw attention of organic chemists due to their various biological activities such as antithrombotic agents1 , antitumor activity[2] , antinociceptive activity[3] , anti-inflammatory

activity4 , anti-oxidants5 , antibacteria[l6] , antifunga[l7] , nematicidal agents8 , antifungal[9] , anti-viral10, anti inflammatory and hypo glycemic agents[11] . On the other hand thiazoles are basic class of heterocyclic moieties which possess a wide range of therapeutic interest and their importance is also very much-established in medicine[12] such as antibacterial and antifungal activities[13-14], ant tubercular activity[15-16], anti HIV agents[17]. Among the important heterocyclic compounds 1,3,4-thiadiazoles are one of the important structural fragments in medicinal chemistry due to their various biological activities, such as Ca^{+2} channel blockers[18], antiinflammatory active agents[18] antitubercular activity, antiinfective agents, antibacterial, antidepressants, anti cancer agents. In view of the above observations it was a thought of interest to design and synthesize a new class of isoxazolopyrazolone derivatives incorporating with 1,3,4-thiadiazole moiety. So, in this present communication we report a facile synthesis of diverse (2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl)-N-(((S)-1-(4-acetyl-4,5-dihy dro-5-methyl-5-phenyl-1,3,oxadiazolyl)ethylcarbamoyl) met hy l)] acetamide derivatives and their antimicrobial activities against various organisms.

Meterials and methods

Thin layer chromatography was used to monitor the completion of the reaction and homogeneity of the synthesised compound. Melting points were determined in open capillary tubes and uncorrected. I.R spectra in KBr pellets were recorded Perkin-Elmer Spectrum 100 FT IR spectrometer (400 MHZ)in DMSO-d6 /CDCl3 using TMS as an internal standard (chemical shifts are expressed in ppm). The homogeneity of the compounds was checked on silica gel–G coated plates by using chloroform and ethylacetate (5:2) as the eluent and observed in U.V light. All the synthesised compounds gave satisfactory elemental analysis

Reagents and condition

Reagents and conditions: (a) Sodium nitrite, Con HCl, 0^oC; (b) Pentane 2,4-dione, CH_3COONa, ethanol, 0^oC; (c) hydrazine hydrate, DMF, MW, Basic Al_2O_3, 150W; (d) Ethyl 2-chloro acetate, Anhydrous K_2CO_3, DMF, RT, 8h; (e) 5N NaOH, THF, RT, 4h; (46) (i) N-Boc Glycine, Alanine methylester. HCl, NMM, DCC, $CHCl_3$, 0^oC; (ii) TFA, MDC; (f) NMM, DCC, $CHCl_3$, 0^oC; (g) hydrazine hydrate, ethanol, (49) (i) Aniline, chloral hydrate, Na_2SO_4, Con. HCl,

Hydroxylamine.HCl, water; (ii) H_2SO_4, 80^oC, 30 min; (h) glacial acetic acid, Methanol, (i) acetic anhydride, reflux, 2h.

Experimental section:

I-synthesis of (2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl) acetamido ethan am id o]-N'-(1-phenylethylidene) propyl hydrazide 1a.

To solution of (2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl)acetamido ethanamido] propyl hydrazide (0.01 mole) in hot methanol (25ml), acetophenone (0.01 mole) and a drop of gl. acetic acid were added. The solid that separated on refluxing for 3 hours was filtered wash with cold methanol and recrystalized from methanol to give (2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl)acetamidoethanamido]-N'-(1-phenylethylidene) propyl hydrazide a. m.p 254^0C, yield 84%. The similar procedure was adopted for the synthesis of 1(b-j) from (b-f).The above reaction of 1a-j with acetophenone has been extended to *p*-methyl acetophenone, *p*-chloroacetophenone, *p*-bromoacetophenone and *p*-nitroacetophenone. The similar procedure was adopted for the synthesis of 1(b-j) from (b-f). The compounds synthesized 1a-j has been characterized by their elemental analysis IR, ^{1}HNMR, and MS data.

IR spectra:The IR (KBr) spectra of (2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl) acetamido ethanamido]-N'-(1-phenylethylidene)propyl hydrazide a showed absorptions around. 3185(m, -NH, str, hydrazine), 2954-2925(m, -CH str, asym, -CH_3 and -CH_2), 2852(m, -CH_2 str, sym, -CH_3), 2210, (s, -N=N str, Azo), 1710, 1645 & 1636 (s, >C=O str, 2^o amides), 1600(s, -C=N- str,(2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl)acetamidoethanamido]-N'-(1-phenylethylidene)propyl hydrazide (a-j).hydrazine), 1540 (m, -NH- bend, 2^o amide), 1460 and 1455 cm^{-1} due to five member hetero cyclic pyrazole ring respectively;

NMR spectra: ^{1}H The ^{1}H NMR spectra of (2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl) acetamidoethanamido]-N'-(1-phenylethylidene) propylhydrazide a was recorded in ^{1}HNMR (200MHz) ($CDCl_3$+DMSO-d_6) (δppm): δ 6.62-8.09(m, 10H, Ar-H), 6.22(br,s, 3H, -NH, amide), 4.86-4.8(1H, m, α-H, Ala), 4.67(S, 2H, CH_2, amide), 3.49-3.47(S, 2H, CH_2, Gly), 2.8(s, 6H,(CH_3)$_2$, pyrazol), 2.1(s, 3H, -CH_3 of phenylethylidene), 1.29-1.27(3H, d, -CH_3, β-H's of Ala).

Massspectra:-(2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl) acetamido ethanam ido]-N'-(1-phenylethylidene) propyl hydrazide a (R = H, R_1 = CH_3, R_2 = C_6H_5) showed molecular ion (M^+) peak at m/z 376.

II-Synthesis of 1,3,4-oxadiazole derivatives containing 3,5-dimethyl pyrazole moiety.

Synthesis of (2R)-[2-(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl)-N-(((S)-1-(4-acetyl-4, 5 dihy dro-5-methyl-5-phenyl-1,3,4-oxadiazolyl)ethylcarbamoyl) methyl)]acetamide(2a).A mixtu re of (2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl) acetamido ethanamido]-N'-(1-p henylethylidene) propyl hydrazide (a) (0.01 mole) and excessive acetic anhydride (10 ml) was refluxed for 2 hours. The excessive acetic anhydride was distilled off and the residue was poured on to crushed ice. The solid thus obtained was filtered, washed with water and recrystalized from aq. methanol to furnish (2R)-[2-(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl)-N-(((S)-1-(4-acetyl-4,5dihydro-5-methyl-5-phenyl-1,3,4-oxadiazolyl)ethyl carbamoyl) me t hyl)]acetamide (2a), m.p. 260^0C, yield 56%. The similar procedure was adopted for the synthesis of 2(b-j) from 1(b-j). The analytical data of 2(b-j) are shown in below.

The cyclization reaction was extended to other hydrazones 2b–j and in each case the respective (substituted) R_2 = *p*-$CH_3C_6H_4$, *p*-ClC_6H_4, *p*-$OCH_3C_6H_4$, *p*-$NO_2C_6H_4$ was isolated in 57 – 68% yields (Table VI). The similar procedure was adopted for the synthesis of 72 (b-f) from 71(b-f). The compounds synthesized 2a-j have been characterized by means of their elemental analysis IR, ^{1}HNMR, and MS data.

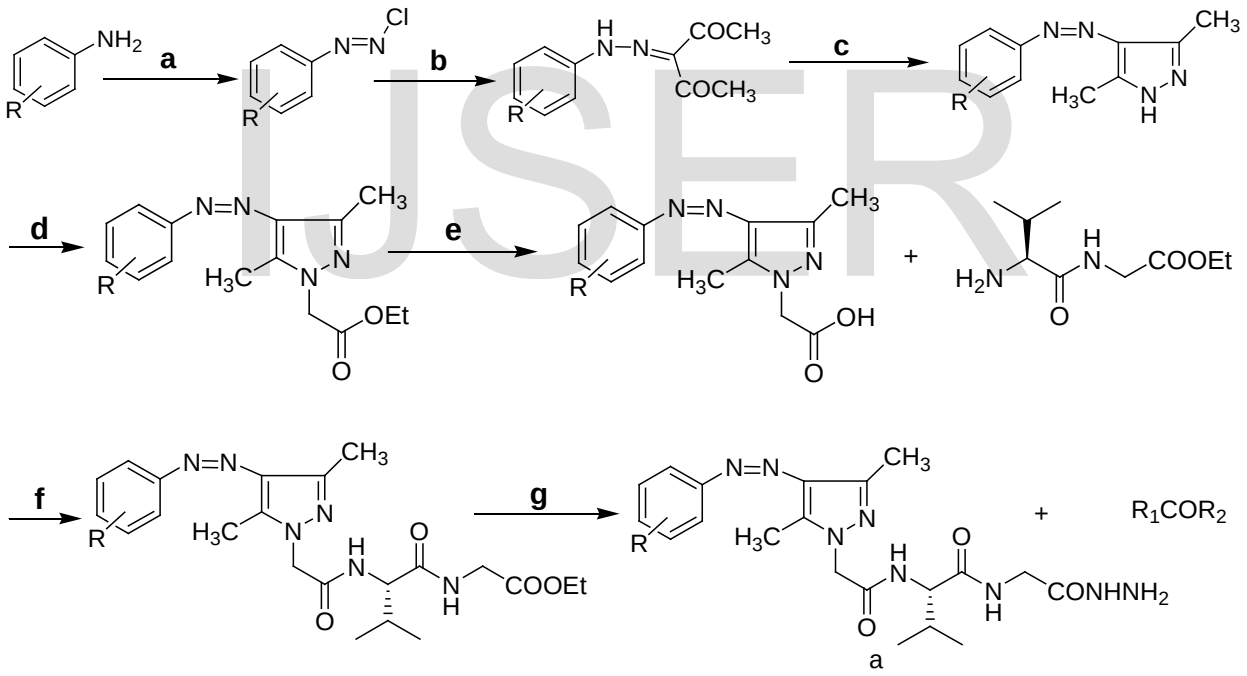
IR spectra:The IR (KBr) spectra of 2(R)-[2-(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl)-N-(((S)-1-(4-acetyl-4,5-dihydro-5-methyl-5-phenyl-1,3,4-oxadiazolyl)ethylcarbamoyl)methyl)] acetamide 72a-f showed absorptions around 3205 (m, -NH, str,), 2954-2925(m, -CH str, asym, -CH_3 and -CH_2), 2852(m, -CH_2 str, sym, CH_3), 2210, (s, -N=N str, Azo), 1710 & 1645 (s, -C=O str, 2^o amides), 1750 (s, N-C=O-CH_3 str, 3^o amides), 1620(s, C=N str, hydrazine), 1545 (m, -NH bend, 2^o amide), 1460 and 1455 cm^{-1} due to five member hetero cyclic pyrazolering respectively;

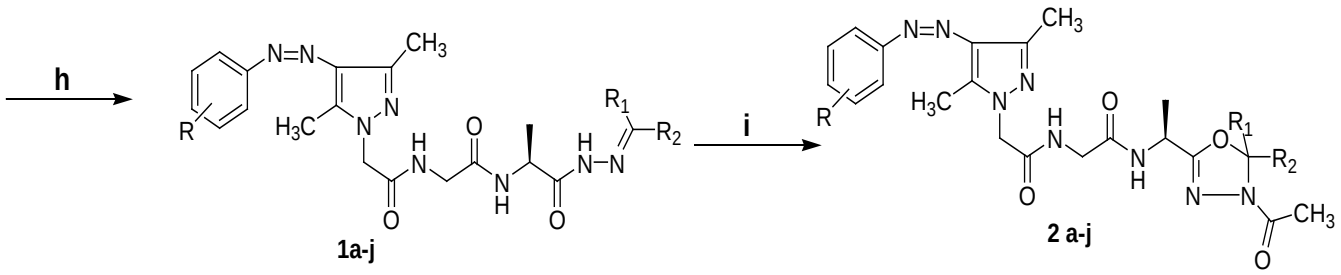
^{1}HNMR spectra; The ^{1}H NMR spectra of 2(R)-[2-(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl)-N-(((S)-1-(4-acetyl-4,5-dihydro-5-methyl-5-phenyl-1,3,4 oxadiazolyl)ethylCarbamoyl) me thyl)]acetamide 2a was recorded in ^{1}HNMR (200MHz) ($CDCl_3$+DMSO-d_6) (δppm): δ 6.68-

8.09(m, 10H, Ar-H), 6.22(br,s, 2H, -NH, amide), 5.42-5.39(1H, q, α-H, Ala), 4.67(S, 2H, CH_2, amide), 3.49-3.47(S, 2H, CH_2, Gly), 2.8(S, 6H, $(CH_3)_2$, pyrazol), 2.1(s, 3H, -CH_3 of phenyl ethylidene), 1.56-1.52(3H, d, -CH_3, β-H's of Ala).

Mass spectra;-The mass spectra of 2,4-(Acetyl-5-methyl-5-phenyl-4,5-dihydro-[1,3,4] oxadiazol-2-methyl)-3,5-dimethyl-4-(phenyl hydrazono)-2,4-dihydro-pyrozole 73a (R = H, R_1 = CH_3, R_2 = C_6H_5) displayed molecular ion (M^+) at m/z 418.The spectrum showed the molecular ion (M^+) peak at m/z 418 with an intensity of 11.4 percent, decomposition of molecular ion A at path-a resulted in the formation of fragment B at m/z 375 (3.2%). Loss of CH_3CHO radical from A yield cation B. Elimination of C_6H_5 radical by path –b from molecular ion A lead to the occurrence of cation m/z 341 (22.2%) and F at m/z 121 (100%) respectively.

Synthetic Root for target molecule;-





Biological activity.

Antibacterial activity and antifungal activity of synthesized compounds

The compounds were tested in-vitro for their antibacterial activity against two microorganisms viz. Escherichia coli and Staphylococcus aureus, which are pathogenic in human beings by cup-plate agar diffusion method. The compounds were tested in vitrofor their antifungal activity against Aspergillus oryzae and Aspergillus niger by cup-plate agar diffusion method.

Procedure: All compounds were screened for antibacterial activity against E. coli and S. aureus by cup plate method For anti bacterial activity, we had taken 20 gm of luria broth (Hi media M-575) and 25 gm of agar–agar in 1000 ml distilled water and heated till it dissolved. Then, the mixture was sterilized by autoclaving at 15 lbs pressure and 121 °C for 15 min. Here, agar–agar was used to solidify the solution. After that, six Petri dishes having flat bottom were taken and filled with about 18 ml of the above solution. Overlay the plate with 4 ml soft agar–agar containing 0.1 ml test culture. Bored four well of 8 mm diameter in each plate. We had then dissolved the compound in DMF having 1000 ppm concentration and added 0.1 ml of testing solution into each well. This solution was allowed to diffuse at 4 °C. After 20 min of diffusion, the plate was incubated at 37 °C overnight. After incubation, we observed the zone of inhibition and measured the diameter of the zone. For anti fungal activity, we had taken 20 gm Sabouraud dextrose instead of lubria broth and followed the same procedure as above. All the synthesized compounds showed good antimicrobial activity

Antibacterial activity and antifungal activity of synthesized compounds

	Anti bacterial blank 12m m	Anti fungal blank 11mm

s.no	E.Coli	S.aureus	A.niger	A.oryzae
Ia	12.00	12.25	9.25	9.25
Ib	12.25	12.50	9.25	9.50
Ic	12.25	11.50	9.25	9.50
Id	13.25	11.25	9.00	9.25
Ie	13.25	13.25	10.00	10.25
If	12.50	12.20	9.25	9.25
Ig	11.50	11.25	9.25	9.00
Ih	11.75	12.20	9.50	9.00

Results and discussion:-

The main aim of this work was to synthesize various substituted (2R)-[2-(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl)-N-(((S)-1-(4-acetyl-4,5dihydro-5-methyl-5-phenyl-1,3,4-oxadiazolyl)ethyl carbamoyl) methyl)]acetamide derivatives Initially were synthesized by (2R)-2-[(4-phenyldiazenyl-3,5-dimethyl-1H-pyrazol-1-yl) acetamido ethan am id o]-N'-(1-phenylethylidene) propyl hydrazide by using different compounds..

All the synthesized compounds resulted in good yields with 50-65%. The formation of title compounds (Ia-1j) is indicated by the IR and ^{1}H-NMR spectrum as given above. The IR of these compounds showed the presence of peaks due to (C=N and C-O-C) of the oxadiazole ring in all the compounds (Ia-Ij). The mass spectra of the title compounds are in confirmative with the assigned structure. The mass spectrum of these compounds showed molecular ion peaks corresponding to their molecular formula Among the newly synthesized compounds showed good antifungal activity. showed better antibacterial activity.Ia,-Ij

Conclusion

From the above review it is evident that oxadiazole and their heterocyclic derivatives represent a promising class of compounds as they show a wide range of pharmacological activities, such as: anti-, antimicrobial, antifungal, antibacterial A number of substitutions are possible in the aromatic rings and ring closure reactions of the oxadizoles affords heterocyclic derivatives which are pharmacologically active which further warrants the exploitation of this class of compounds.

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