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Research article

Synthesis of reserpine coupled 1,3,4-oxadiazole derivatives and their biological evaluations

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Key words: Reserpine, 1,3,4-Oxadiazole, Anti-inflammatory activity; Antibacterial activity.

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.

Abstract

Some novel reserpine coupled 1,3,4-oxadiazole derivatives (RAH_1-RAH_{12}) were synthesized in good yields and evaluated for their possible antibacterial, anti-inflammatory activities. The structures of the synthesized compounds were confirmed on the basis of their spectral data and elemental analysis. Their antibacterial activities were evaluated by the disc diffusion method while their anti-inflammatory activities were evaluated by the carrageenan-induced rat paw edema test. All the tested compounds showed moderate to good antibacterial activity when compared with chloramphenicol and high to moderate anti-inflammatory activity when compared to indomethacin. The obtained results showed that the most active compounds could be useful as a template for future design, modification and investigation to produce more active analogs.

Introduction

The development of an effective therapeutic agent for the management of inflammation has undergone continual evolution leading to the emergence of more efficacious classes of drugs. Since the discovery of aspirin, much efforts have been devoted to the development of non-steroidal anti-inflammatory drugs (NSAIDs), which are among the most widely prescribed medication in clinical practice despite their documented renal and gastrointestinal (GI) side effects. Conventional NSAIDs exert non-selective inhibition [1] of COX enzymes, the agents which catalyze the rate-limiting step in the formation of prostanoids from arachidonic acid. Such indiscriminate inhibition of COX-1 as well as COX-2 has been blamed for high incidence of GI irritation or, in the worst case, development of life threatening GI ulcers and bleeding in long term users of NSAIDs. Consequently, a second generation of NSAIDs has been developed which selectively inhibit COX-2. Being selective COX-2 inhibitors, these are expected to achieve the same anti-inflammatory efficacy as traditional NSAIDs but minimize the risk of unwanted GI complications. Though the selective COX-2 inhibitors have minimal toxicity in the gastrointestinal tract, these agents can produce severe side effects in renal, hepatic, and cardiovascular systems. The recent withdrawal of valdecoxib [2] and rofecoxib [3] has focused attention on the adverse cardiovascular effects of selective COX-2 inhibitors [4]. Thus, search for novel anti-inflammatory drugs with minimal GI side effects and high safety margin is still warranted.

Discovery of potent and effective antibacterial agents represents one of the most important advances in therapeutics, not only in the control of serious infections, but also in the prevention and treatment of some infectious complications of other therapeutic modalities such as cancer chemotherapy and surgery. Over the past decade, microbial infection becomes an important complication and a major cause of morbidity and mortality in immuno-compromised individuals such as those suffering from tuberculosis, cancer and AIDS and in organ transplantation cases [5, 6]. Despite the availability of large number of antibacterial drugs, the increase in diseases attributed to drug-resistant and multi-drug resistant (MDR) bacterial pathogens indicates a growing need for new antibacterial drugs. The most difficult types of infections to treat are those involving Gram-negative bacteria and mycobacteria, since these classes of bacteria often possess multiple mechanisms that make them resistant to entire classes of antibacterial drugs [7]. Consequently, the development of newer antimicrobial agents will remain an important challenging task for medicinal chemists.

It is reported that reserpine is having varied biological activity such as antihypertensive, sedative & hypnotics, tranquilizer, cure pain due to affection of bowels, in a labour to increase uterine contraction, asthma and dermatological disorders[8, 9]. Therefore, we have selected reserpine as a test molecule and prepared its semisynthetic analogues having hydrazide and 1,3,4-oxadiazole nucleus. 1,3,4- oxadiazole derivatives have been explored for interesting pharmacological properties

like anti-tubercular, bacteriostatic, hypoglycemic, diuretic, anti-inflammatory, anti-viral, anti-fungal, sedative and hypnotic activity [10-13].

Till date the derivatives of reserpine having 1, 3, 4-oxadiazole nucleus has not been reported. Therefore it is thought worthwhile to prepare semisynthetic analogues of reserpine having 1, 3, 4-oxadiazole nucleus. The synthesized compounds have been characterized by various spectral analysis and these synthesized compounds were screened for anti-inflammatory and antibacterial activities.

Materials and Methods

Chemistry

All the starting materials and solvents used in the synthesis were obtained from Himedia Laboratories, Mumbai and S.D. Fine Chemicals and used without further purification. The homogeneity of the compounds was monitored by ascending thin layer chromatography (TLC) on silica gel-G (Merck) coated aluminum plates, visualized by iodine vapour/UV chamber. The melting points were determined in open capillary tubes. FT-IR 8400 Shimadzu Spectrometer (IR analysis), Waters Q-T of Micromass (ESI-MS) and Thermo Finnigan (elemental analysis) were used for analysis.

General Procedure for synthesis of (RAH_1- RAH_{12})

Solution of reserpic acid hydrazide (0.01 mole) in phosphorous oxychloride (3-5 mL) and several aromatic acid (0.01 mol) were added in the round bottom flask followed by refluxed for 5-8 hr. After completion of reaction (monitored by TLC), cooled to RT, mixture was neutralized with ice cold solution of 10% $NaHCO_3$, a solid mass obtained was filtered and washed using ice cold water and crystallized from suitable solvents. The purity of synthesized compounds was checked by TLC.

2,11-Dimethoxy-1-(5-phenyl-[1,3,4]oxadiazol-2-yl)-1,2,3,4,4a,5,7,8,13,13b,14,14a-dodecahydro-indolo[2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_1)

Nature, Brown solid; M.P. 316 ^{0}C (decomposed); Yield: 71%; Mol. Formula: $C_{29}H_{32}N_4O_4$; Mol. Weight: 500.59; IR (KBr, cm^{-1}): 3415 (OH), 3203 (NH str), 3068 (Ar-CH), 1556 (C=N), 1118, 1067,989 (C-O-C) M/S 501.40 $[M+1]^+$; % Anal. Cal. for $C_{29}H_{32}N_4O_4$; C, 69.58; N, 11.19; H, 6.44; Found: C, 60.445; N, 9.908; H, 5.332.

1-[5-(4-Bromophenyl)-[1,3,4]oxadiazol-2-yl]-2,11-dimethoxy-1,2,3,4,4a,5,7,8,13,13b,14, 14a-dodecahydroindolo[2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_2)

Nature, Brown solid; M.P.280^{0}C (decomposed); Yield: 74 %; Mol. Formula: $C_{29}H_{31}BrN_4O_4$; Mol. Weight: 578.15; IR (KBr, cm^{-1}): 3303 (NH str), 3225(OH), 2938 (Ar-CH),1631 (C=N), 1158, 1072, 1010 (C-O-C), M/S 579.58 $[M+1]^+$; % Anal. Cal. for $C_{29}H_{31}BrN_4O_4$; C, 60.11; N, 9.67; H, 5.39; Found: C, 60.445; N, 9.908; H, 5.332.

1-[5-(4-Nitrophenyl)-[1,3,4]oxadiazol-2-yl]-2,11-dimethoxy-1,2,3,4,4a,5,7,8,13,13b,14, 14a-dodecahydroindolo[2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_3)

Nature, Blackish brown solid; M.P. 360^{0}C (decomposed); Yield: 79%; Mol. Formula: $C_{29}H_{31}N_5O_6$; Mol. Weight: 545.23; IR (KBr, cm^{-1}): 3441 (NH str), 3374 (OH), 3126 (Ar-CH), 1709 (C=N), 1578 (NO_2), 1136, 1120, 1067, 993 (C-O-C); M/S5 46.58 $[M+1]^+$; % Anal. Cal. for $C_{29}H_{31}N_5O_6$; C, 63.84; N, 12.84; H, 5.73; Found: C, 62.565; N, 11.998; H, 6.5.117.

1-[5-(4-Chloro-phenyl)-[1,3,4]oxadiazol-2-yl]-2,11-dimethoxy-1,2,3,4,4a,5,7,8,13,13b,14 ,14a-dodecahydroindolo[2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_4)

Nature: Dark brown powder; M.P. 296^{0}C (decomposed); Yield: 86%; Mol. Formula: $C_{29}H_{31}ClN_4O_4$; Mol. Weight: 534.20; IR (KBr, cm^{-1}): 3219 (NH str), 2938 (OH), 2843 (Ar-CH), 1631 (C=N), 1157, 1092, 1013 (C-O-C), 739 (C-Cl); M/S 535.43 $[M+1]^+$; % Anal. Cal. for $C_{29}H_{31}ClN_4O_4$; C, 65.10; N, 10.47; H, 5.84; Found: C, 65.790; N, 10.868; H, 5.587.

1-[5-(P-tolyl)-[1,3,4]oxadiazol-2-yl]-2,11-dimethoxy-1,2,3,4,4a,5,7,8,13,13b,14,14a-dodecahydroindolo [2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_5)

Nature: Brown solid; M.P. 330^{0}C (decomposed); Yield:56%; Mol. Formula: $C_{30}H_{34}N_4O_4$; Mol. Weight: 514.26; IR (KBr, cm^{-1}): 3240 (NH str), 2940 (Ar-CH), 1630 (C=N),1467(C=C), 1160, 1088, 912 (C-O-C), 763 (C-H bending);M/S 515.49 $[M+1]^+$; % Anal. Cal. for $C_{30}H_{34}N_4O_4$; C, 70.02; N, 10.89; H, 6.66; Found: C, 70.340; N, 10.675; H, 6.989.

1-[5-(2-Hydroxy-phenyl)-[1,3,4]oxadiazol-2-yl]-2,11-dimethoxy-1,2,3,4,4a,5,7,8,13, 13b,14, 14a-dodecahydroindolo[2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_6)

Nature: Dark brown solid; M.P. 346^{0}C (decomposed); Yield: 72%; Mol. Formula: $C_{29}H_{32}N_4O_5$; Mol. Weight:516.24; IR (KBr, cm^{-1}): 3326(NH), 3240 (OH str), 2942 (Ar-CH), 1630 (C=N), 1477(C=C); 1344 (OH bending), 1104, 980 (C-O-C), 769 (C-H bending); M/S 517.52 $[M+1]^+$; % Anal. Cal. for $C_{29}H_{32}N_4O_5$; C, 67.43; N, 10.85; H, 6.24; Found: C,67.670; N, 11.108; H, 6.188.

1-[5-(3-Nitro-phenyl)-[1,3,4]oxadiazol-2-yl]-2,11-dimethoxy-1,2,3,4,4a,5,7,8,13,13b,14, 14a-dodecahydroindolo[2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_7)

Nature: brown solid; M.P. 292^{0}C (decomposed); Yield: 75%; Mol. Formula: $C_{29}H_{31}N_5O_6$; Mol. Weight: 545.59; IR (KBr, cm^{-1}): 3444 (OH str), 3369 (NH), 3076 (Ar-CH), 1658 (C=N), 1556 (NO_2) ,1408 (C=C), 1346 (NO_2), 1118, 989, 948 (C-O-C); M/S 546.54 $[M+1]^+$; % Anal. Cal. for $C_{29}H_{31}N_5O_6$; C, 63.84; N, 12.84; H, 5.73; Found; C, 62.433; N, 12.932; H, 5.142.

1-[5-(2-Bromo-phenyl)-[1,3,4]oxadiazol-2-yl]-2,11-dimethoxy-1,2,3,4,4a,5,7,8,13,13b,14, 14a-dodecahydroindolo[2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_8)

Nature: Dark brown solid; M.P. 296^{0}C (decomposed); Yield: 79%; Mol. Formula: $C_{29}H_{31}BrN_4O_4$; Mol. Weight: 578.15; IR (KBr, cm^{-1}): 3284 (OH str), 3219 (NH), 2981 (Ar-CH), 1620 (C=N), 1423(C=C), 1199, 1134, 1072, 1034 (C-O-C), 865 (Ph), 651 (C-Br) Cm^{-1}; M/S 579.57 $[M+1]^+$; % Anal. Cal. for $C_{29}H_{31}BrN_4O_4$; C, 60.11; N, 9.67; H, 5.39; Found: C, 60.580; N, 9.117; H, 5.098.

1-[5-(3-Bromo-phenyl)-[1,3,4]oxadiazol-2-yl]-2,11-dimethoxy1,2,3,4,4a, 5,7,8,13,13b,14,14a-dodecahydroindolo[2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_9)

Nature: Yellowish brown solid; M.P. 297^{0}C (decomposed); Yield: 71%; Mol. Formula: $C_{29}H_{31}BrN_4O_4$; Mol. Weight: 578.15; IR (KBr, cm^{-1}): 3437 (OH str), 3143 (NH), 2981 (Ar-CH), 1664,1558 (C=N), 1415(C=C), 1120, 991 (C-O-C); M/S 579.50 $[M+1]^+$; % Anal. Cal. for $C_{29}H_{31}BrN_4O_4$; C, 60.11; N, 9.67; H, 5.39; Found: C, 60.250; N, 9.248; H, 5.757.

1-[5-(Styryl)-[1,3,4]oxadiazol-2-yl]-2,11-dimethoxy-1,2,3,4,4a,5,7,8,13,13b,14,14a-dodecahydroindolo [2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_{10})

Nature: Dark brown solid; M.P. 326 ^{0}C (decomposed); Yield: 73%; Mol. Formula: $C_{30}H_{32}N_4O_4$; Mol. Weight: 512.24; IR (KBr, cm^{-1}): 3229 (NH), 2941 (Ar-CH), 1621, 1573 (C=N), 1460(C=C), 1163, 1082, 929 (C-O-C), 826 (Ph); M/S 513.66 $[M+1]^+$; % Anal. Cal. for $C_{30}H_{32}N_4O_4$; C, 70.70; N, 10.64; H, 6.51; Found: C, 70.389; N, 10.255; H, 6.899.

1-[5-(3,5-Dinitro-phenyl)-[1,3,4]oxadiazol-2-yl]-2,11-dimethoxy-1,2,3,4,4a,5,7,8,13,13b, 14,14a-dodecahydroindolo[2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_{11})

Nature: Blackish brown solid; M.P.350 ^{0}C (decomposed); Yield: 65%; Mol. Formula: $C_{29}H_{30}N_6O_8$; Mol. Weight: 590.21; IR (KBr, cm^{-1}): 3379 (OH), 3298 (NH), 2981 (Ar-CH), 1600 (C=N), 1336 (NO_2), 1168 (C-O-C str. oxadiazole), 1168 (C-O-C str. oxadiazole), 1066 (N-N str. oxadiazole), 952 (C-O-C), 868 (Ph); M/S 591.57$[M+1]^+$; % Anal. Cal. for $C_{29}H_{30}N_6O_8$; C, 58.98; N, 14.23; H, 5.12; Found: C, 57.230; N, 15.852; H, 5.128.

1-[5-(Furan-2-yl)-[1,3,4]oxadiazol-2-yl]-2,11-dimethoxy-1,2,3,4,4a ,5,7,8, 13,13b,14,14a-dodeca hydroindolo[2',3':3,4]pyrido[1,2-b]isoquinolin-3-ol (RAH_{12})

Nature: Yellowish brown solid; M.P. 310 ^{0}C (decomposed); Yield: 77%; Mol. Formula: $C_{27}H_{30}N_4O_5$; Mol. Weight: 490.55; IR (KBr, cm^{-1}): 3348 (OH), 3223 (NH), 2993 (Ar-CH), 1647 (C=N), 1568 (NO_2), 1116, 1020, 1053, 987 (C-O-C oxadiazole & furan), 864 (Ph); M/S 490.55 $[M+1]^+$; % Anal. Cal. for $C_{27}H_{30}N_4O_5$; C, 66.11; N, 11.42; H, 6.16; Found: C, 65.175; N, 11.258; H, 6.257.

Pharmacological Study

Anti-inflammatory activity

Wistar rats of one of either sex (180–200 g) were used excluding pregnant. In one group 6 animals are present. The animals were kept in RT and food and water were given to acclimatized for 2 days. This activity was done by the method described by winter *et al*[14] (paw oedema method). The tests were evaluated by studying inflammatory response in the rats by injecting Carrageenan, which produces edema. The compounds were evaluated by examining their ability to diminish or prevent the edema. Indomethacin was used as standard drug for this study.

Antibacterial activity

Nutrient agar and Vogel Johnson agar plates were prepared by pouring 15-20 ml of the medium into each sterilized Petri dishes (washed and dried in hot air oven at 160^0 C for 1 hour) and were allowed to solidify at room temp (RT). The medium was seeded with organism by spread plate method using sterile cotton swabs. Three cups were scooped in each plate using a sterile cork borer of 8 mm diameter. Then the solution of test compounds (0.06 ml, 0.08 ml and 0.10 ml) was added to the respective bores by using micropipettes. 0.06 ml, 0.08 ml and 0.10 ml of the chloramphenicol were taken as a standard reference. DMSO was used as control. The Petri dishes were incubated at 37 ^{0}C for 24 h and zone-inhibition was observed and measured using scale (in mm) for each organism [15, 16]. *E. coli* and *S. aureus* were used as microorganism for testing of the synthesized compounds. The media was used for both sub-culturing and also for estimating antibacterial activity. Chloramphenicol was used as standard for antibacterial activity.

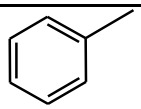
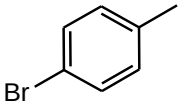
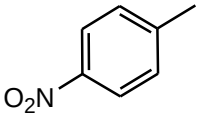
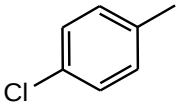
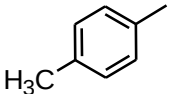
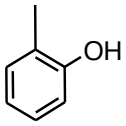
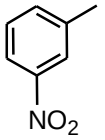
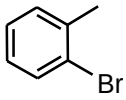
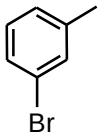
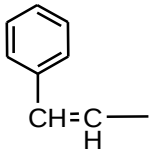
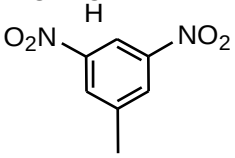
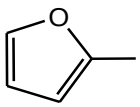
Result and Discussion

Chemistry

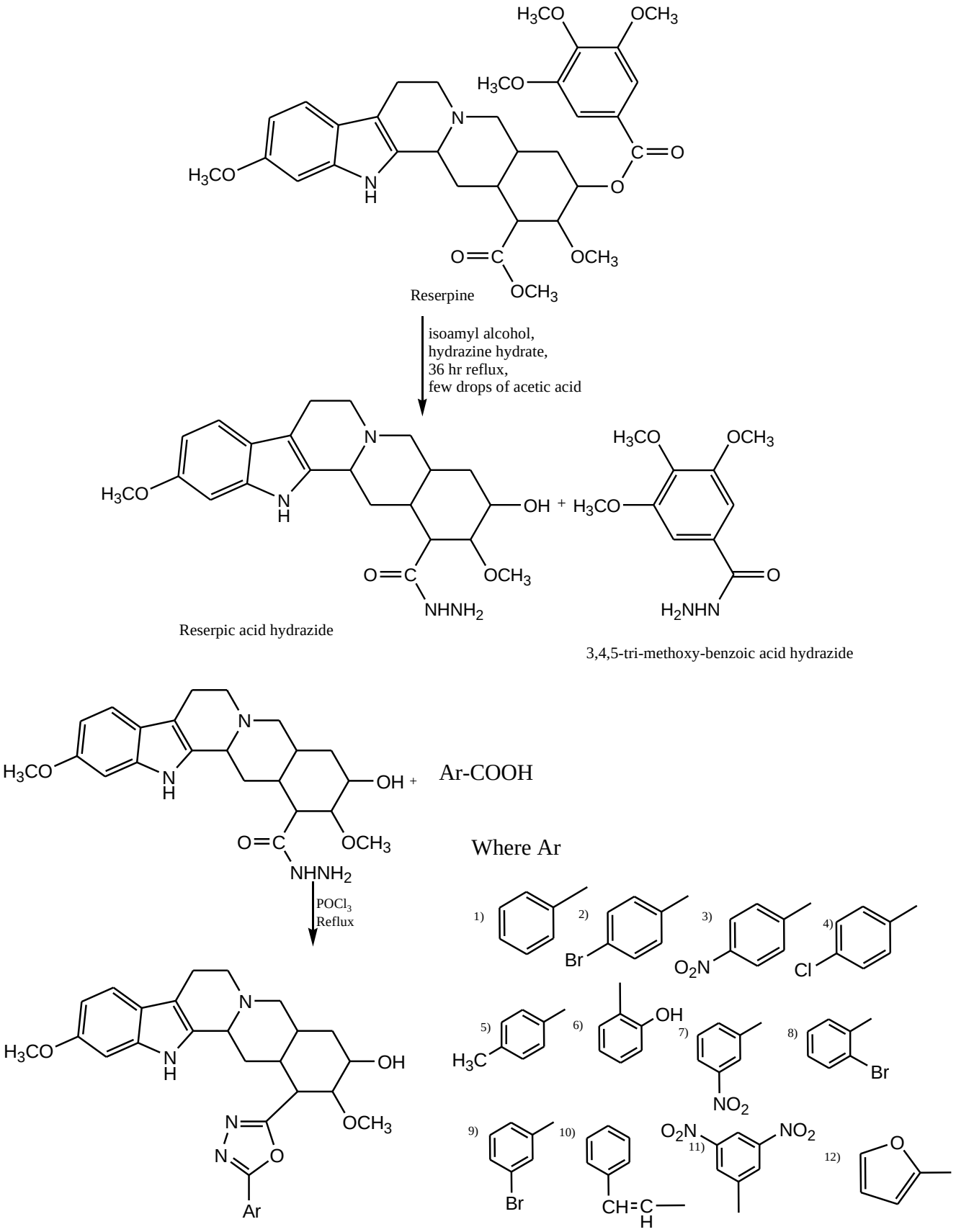
Reserpic acid hydrazide **(RAH)** was synthesized by reacting reserpine with hydrazine hydrate as per reported method [17]. We expanded our series and synthesized twelve 1,3,4-oxadiazole derivatives of reserpic acid hydrazide **(RAH_1-RAH_{12})**. The compounds **RAH_1-RAH_{12}** were obtained by reacting compound RAH with

various substituted aromatic acids (Scheme 1). The isolated yields of synthesized compounds were in the range of 71- 86 % and reactions were completed in about 5-8 hr (monitored by TLC). The compounds were purified by recrytallization and purity was checked by TLC method. Melting points were determined in open capillary tubes. The physical data for the compounds are presented in Table 1. The formation of synthesized compounds was confirmed by means of IR, Mass and elemental analysis spectroscopy and data suggested for proposed structures.

Table 1. % Yield and physicochemical properties of synthesized compounds (RAH) & (RAH_1-RAH_{12})

Compounds	-Ar	Yield (%)	M. P. (^{0}C)	Molecular formula	Mol. wt.	R_f value
RAH_1		71	316^{0}C (decomposed)	$C_{29}H_{32}N_4O_4$	500.59	0.53 (E:H; 4:1)
RAH_2		74	280^{0}C (decomposed)	$C_{29}H_{31}BrN_4O_4$	578.15	0.67 (E:H; 1.5:3.5)
RAH_3		79	360^{0}C (decomposed)	$C_{29}H_{31}N_5O_6$	545.23	0.57 (E:H; 1.5:3.5)
RAH_4		86	296^{0}C (decomposed)	$C_{29}H_{31}ClN_4O_4$	534.20	0.45 (E:H; 1:4)
RAH_5		56	330^{0}C (decomposed)	$C_{30}H_{34}N_4O_4$	514.26	0.51 (C:M; 3:2)
RAH_6		72	346^{0}C (decomposed)	$C_{29}H_{32}N_4O_5$	516.24	0.77 (E:H; 1.5:3.5)
RAH_7		75	292^{0}C (decomposed)	$C_{29}H_{31}N_5O_6$	545.59	0.75 (E:H; 1:4)
RAH_8		79	296^{0}C (decomposed)	$C_{29}H_{31}BrN_4O_4$	578.15	0.57 (E:H; 1:4)
RAH_9		71	297^{0}C (decomposed)	$C_{29}H_{31}BrN_4O_4$	578.15	0.47 (E:H; 1:4)
RAH_{10}		73	326 ^{0}C	$C_{30}H_{32}N_4O_4$	512.24	0.49 (E:H; 1:4)
RAH_{11}		65	350^{0}C (decomposed)	$C_{29}H_{30}N_6O_8$	590.21	0.60 (E:H; 3:2)
RAH_{12}		77	310^{0}C (decomposed)	$C_{27}H_{30}N_4O_5$	490.55	0.65 (E:H; 1:4)

E: ethyl acetate; H: n-hexane; C: chloroform; M: methanol.



Scheme 1. Synthesis of titled compounds (RAH_1- RAH_{12}).

Pharmacological Study

Antiinflammatory activity

Antiinflammatory activity was carried at time intervals 1hr (Table 2), 2hr (Table 3) and 3hr (Table 4). All compounds have shown anti-inflammatory activity. **RAH_2, RAH_3, RAH_5 and RAH_{10}** showed same activity when compared with standard drug (indomethacin) at the dose of 10 mg/kg after 1 hour. In the second hour of inhibition, compounds **RAH_4, RAH_6, RAH_7, RAH_{11} and RAH_{12}** have shown increased inhibition as compared to the first hour. The remaining compounds have shown decrease in inhibition as compared to the first hour. In the third hour of inhibition, compounds **RAH_2, RAH_3, RAH_7, RAH_8, RAH_9, RAH_{10}** and **RAH_{12}** have shown the increased inhibition as compared to the second hour. The compounds **RAH_7** and **RAH_{11}** were found to show less activity among the derivatives. Overall result of anti-inflammatory activity showed that the reserpine scaffold and 1,3,4-oxadiazole ring seem to be pharmacophore and other substituents may play a role in enhancing the activity. The % inhibition is plotted against the test compound to compare anti-inflammatory with standard indomethacin (Figure 1).

Antibacterial activity

The synthesized compounds (RAH_1- RAH_{12}) have been screened for antibacterial against *E. coli* and *S. aureus*. The synthesized compounds have shown moderate to good antibacterial activity. The compounds RAH_1, RAH_4, RAH_8 and RAH_{10} found to have a very good antibacterial activity for *E. coli* when compared with Chloramphenicol (standard drug). RAH_2 has also shown equipotent activity against *E. coli*. The compound RAH_3 has shown more potent activity than standard for *E. coli*. RAH_8 has shown same activity at dose of 80 µg/ml against *E. coli*. The compounds RAH_2 and RAH_8 have shown comparable activity for *S. aureus* when compared with Chloramphenicol (standard drug). Compound RAH_3 has shown same activity for *S. aureus* when compared with standard drug. Also compound RAH_3 has shown broad spectrum antibacterial activity when compared with standard Chloramphenicol.

Table 2. Mean Paw Volume and % of inhibition of compound after 1 hour.

Substance	0 HOUR	AFTER 1 HOUR	
	Mean-Paw Volume (ml) ± SEM (n=6)	Mean-Paw Volume (ml) ± SEM (n=6)	% inhibition
Control	0.550±0.013**	1.265±0.011**	0
Standard	0.593±0.007**	0.645±0.011**	49.01
RAH_1	0.538±0.008**	0.97±0.011*	23.32
RAH_2	0.549±0.008**	0.755±0.007**	40.31
RAH_3	0.540±0.008**	0.743±0.007**	41.26
RAH_4	0.543±.008**	0.910±0.018**	28.06
RAH_5	0.528±0.006**	0.739±0.006**	41.58
RAH_6	0.545±0.005**	0.970±0.017**	23.32
RAH_7	0.526±0.004**	1.056±0.010**	16.52
RAH_8	0.535±0.006*	0.76±0.006**	39.52
RAH_9	0.530±0.006**	0.86±0.011**	32.00
RAH_{10}	0.542±.007*	0.711±0.008**	43.79
RAH_{11}	0. 535±0.008**	1.103±01008**	12.80
RAH_{12}	0. 549±0.008**	1.128±01008**	10.83

Dose:10 mg/kg, (P< 0.01, *P< 0.05),**

Table 3. Mean Paw Volume and % of inhibition of compound after 2 hour.

Substance	0 HOUR	AFTER 2 HOUR	
	Mean-Paw Volume (ml) ± SEM (n=6)	**Mean-Paw Volume (ml) ± SEM (n=6)**	**% inhibition**
Control	0.550±0.013**	1.372±0.006**	0
Standard	0.593±0.007**	0.742±0.010**	45.91
RAH_1	0.538±0.008**	1.05±0.018**	23.46
RAH_2	0.549±0.008**	1.118±0.012**	18.51
RAH_3	0.540±0.008**	0.920±0.017**	29.31
RAH_4	0.543±.008**	0.87±0.008**	36.58
RAH_5	0.528±0.006**	0.980±0.017**	28.57
RAH_6	0.545±0.005**	0.854±0.006**	37.75
RAH_7	0.526±0.004**	1.12±0.0153**	18.36
RAH_8	0.535±0.006*	1.14 ±0.009**	16.90
RAH_9	0.530±0.006**	0.876±0.013**	36.15
RAH_{10}	0.542±.007*	1.10±0.015**	19.82
RAH_{11}	0. 535±0.008**	0.987±0.009**	28.06
RAH_{12}	0. 549±0.008**	0.970±010**	29.30

Dose:10 mg/kg, (P< 0.01, *P< 0.05)**

Table 4. Mean Paw Volume and % of inhibition of compound after 3 hour.

Substance	**0 HOUR**	**AFTER 3 HOUR**	
	Mean-Paw Volume (ml) ± SEM (n=6)	Mean-Paw Volume (ml) ± SEM (n=6)	% inhibition
Control	0.550±0.013**	1.433±0.008**	0
Standard (10 mg/kg)	0.593±0.007**	0.745±0.082**	48.01
RAH_1	0.538±0.008**	1.130±0.008**	21.14
RAH_2	0.549±0.008**	1.035±0.024**	27.77
RAH_3	0.540±0.008**	0.985±0.020**	31.26
RAH_4	0.543±.008**	0.993±0.010**	30.70
RAH_5	0.528±0.006**	1.130±0.015**	21.11
RAH_6	0.545±0.005**	0.950±014**	33.70
RAH_7	0.526±0.004**	1.140±0.007**	20.44
RAH_8	0.535±0.006*	1.189±0.013**	17.02
RAH_9	0.530±0.006**	0.905.±0.009**	36.84
RAH_{10}	0.542±.007*	0.969±0.010**	32.37
RAH_{11}	0. 535±0.008**	1.047±0.007**	26.93
RAH_{12}	0. 549±0.008**	0.925±010**	35.45

Dose:10 mg/kg, (P< 0.01, *P< 0.05)**

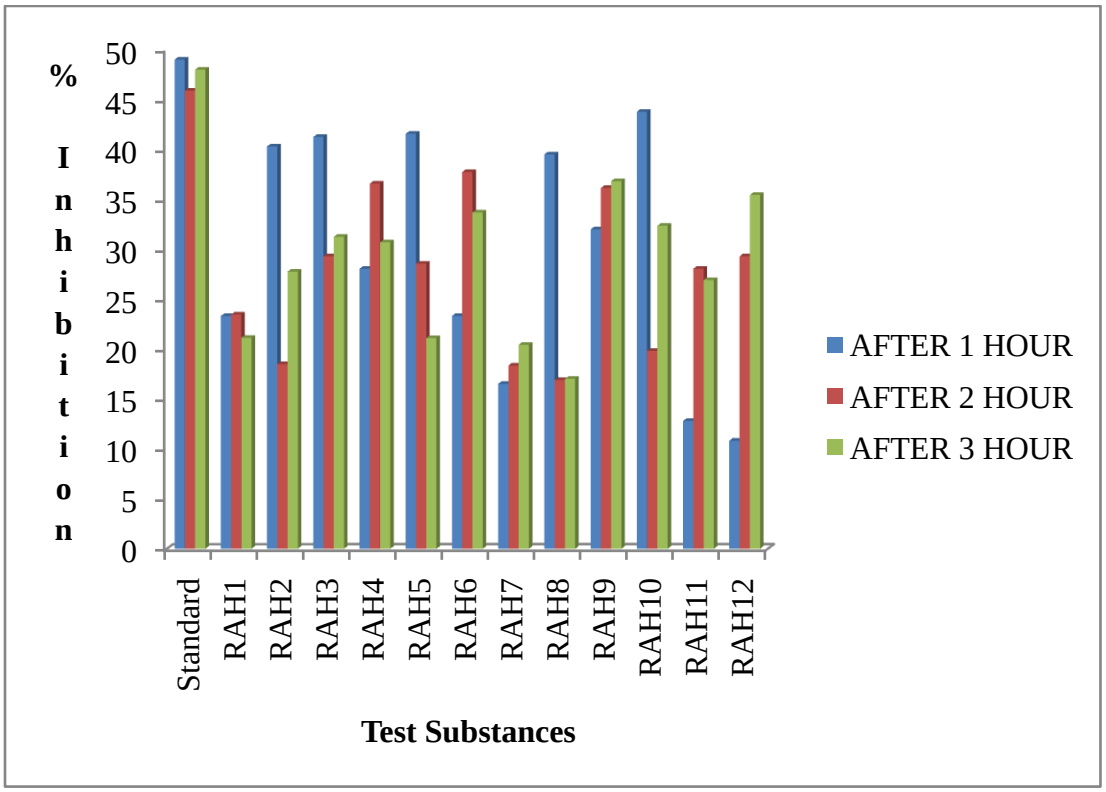
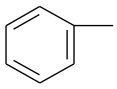
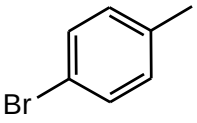
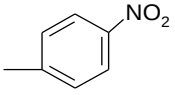
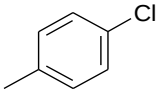
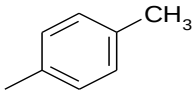
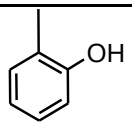
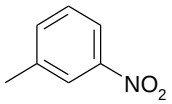
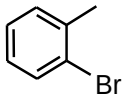
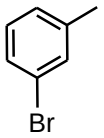
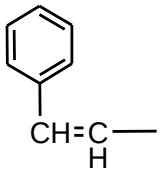
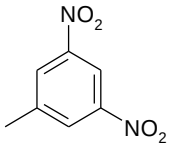
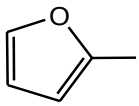


Figure 1. Graph of % of inhibition of compounds RAH_1- RAH_{12}

Table 5. Antibacterial Screening Data of Compound RAH_1- RAH_{12}

Compound No.	Ar	Diameter of Zone Inhibition(in mm)					
		E. coli			*S. aureus*		
		60 µg/ml	80 µg/ml	100 µg/ml	60 µg/ml	80 µg/ml	100 µg/ml
RAH_1		5	7	10	3	5	9
RAH_2		7	0	13	6	9	12
RAH_3		9	12	15	8	1	15
RAH_4		4	6	10	4	5	8
RAH_5		2	3	7	1	3	5

RAH_6		3	5	7	2	4	6
RAH_7		3	5	6	5	7	8
RAH_8		6	9	12	5	8	10
RAH_9		4	5	7	4	7	10
RAH_{10}		5	8	11	4	7	10
RAH_{11}		1	3	5	1	3	6
RAH_{12}		2	5	8	1	2	5
Standard	Chloramphenicol	7	9	13	8	11	14

Conclusion

A series of novel compounds titled **RAH_1-RAH_{12}**were prepared in good yield and purity was checked by TLC. The synthesized compounds structures were established and characterized by spectral analysis like IR, Mass and elemental spectral analysis etc. The synthesized compounds were evaluated for anti-inflammatory activity and compared with standard Indomethacin. Most of the synthesized compounds were activity comparable to standard. The synthesized compounds were also tested for *in vitro* antibacterial activity by disc diffusion method against one Gram-negative microorganism viz., *E. coli* (NCTC 10418), and one Gram-positive microorganism viz., *S. aureus* (NCTC 6571). Chloramphenicol was used as standard drug for antibacterial activity. The synthesized compounds were shown broad spectrum of antibacterial activity. The compounds have shown moderate to good activity. Also compound **RAH_3** has shown broad spectrum antibacterial activity when compared with standard Chloramphenicol. Thus, this research work can be further studied and optimized for development of new antibacterial, and anti-inflammatory agents.

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