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Antibacterial Behavior of New Derivatives of Pyrazinamide on Mycobacterium Tuberculosis

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ABSTRACT

Synthesis, Characterization and antibacterial behavior of some new derivatives of PZA re formed using different acid Chlorides and Sodium Pyrazinamide [Na-PZA]. Synthesis of PZA derivatives was carried out in two steps. A sodium salt of Pyrazinamide was prepared in the first step where as in the second step the final derivatives were prepared using this sodium salt. These derivatives were characterized by their melting points Purity, JR and NMR Spectral data. All derivatives have been secured for their antibacterial behavior against bacteria. This behviours sensitivity test was done on Lowenstein-Jensen medium.

Key Words: Sodium Salt of PZA, Antibacterial behaaviour, Pyrazinamide, Lowenstein – Jensen medium.

INTRODUCTION

Tuberculosis, one of the oldest known human disease, is still one of the major cause of mortality. Tuberculosis is a contagious disease caused in human being by several species of Mycobacterium. It is an organism belonging to the family of mycobacteriacial and the order actinomycetals. It is a slowly growing organism. Tuberculosis most commonly attacks the lungs but virtually every organ of the body may be involved. Streptomycin was established as the first effective antituberculous drug [2]. After a few years, the investigation of hetrocyclic analogs of Nicotinamide [PZA] was discovered by Kushvir [3,4]. Resistance develops rapidly when the drug is given alone [5] while isoniazid and ethionamide may delay resistance, but apparently does not eradicate them completely [6]. Later (Recently) the other antitubucular drugs have been developed such as Isoniazide [7], Rifampicin [8] and ethambutol [9]. However the organism can develop resistance to these drugs therefore the work is still continued to develop a more potent drug as compared to the previous drugs. Hence the present studies are carried out to Synthesis some new derivatives of Pyrazanamide with different acid chlorides and sodium pyrazinamide, as mentioned in the

paper. Which are characterized by elemental analysis and spectral studies and their effect antimycobacterium tuberculosis is studied by Lowenstein-Jensen medium [10, 11] to test these compounds as antitubercular agents.

EXPERIMENTAL WORK

Melting points were measured by capillary method. Elemental analysis of C,H,N;IR spectra and ^{1}HNMR spectra were carried out in research laboratory of CDRI, Lucknow. Purity of compounds was monitored by TLC on Silica gel coated plates.

N-Binzoyl Pyrozinamide (1). Equimolar proportion of Sodium Pyrazinamide in round bottom flask in ice bath and benzoyl chloride solution was then added drop by drop with rigorous continuous stirring and left it over night of room temperature. A light brown coloured compound was formed. The obtained compound was filtered and washed and thereafter recrystallized with alcohol. Yield 8.5 gm. Per 0.5 gm. of Sodium Pyrazinamide, M.P. 100-101°C. Anal. Caled for C12 Hg N3O: C, 63.45; H, 3.96; N, 18.49. Found: C, 62.6; H, 3.68; N, 18.63%; IR: Cm^{-1} 1594.6 [Benzene ring], 1688.6 [C=O], 3062.6 [Aromatic CH-stretching], 3419.8 [NH-Stretching]; ^{1}HNMR 89.297 [Py-C=O]; 7.478 [Ar (C=0)]; 8.702 [O = C-NH-C=O].

N-Salicyl Pyrazinamide (2). The equimolar proportion of sodium Pyrazinamide and Solicyl chloride were mixed and the mixture was heated for very short time. When the reaction appeared to be completed a brown coloured compound was formed. The solid compound formed was filtered washed & recrystallized with alcohol to get dark brown coloured powder. Yield 0.85 gm per 0.5 gm of sodium pyrazinamide, M.P. 130-135^{0}C [Approx]. anal. Calcd. for C_{12}, H_g, N_3 O_3: C, 59.27; H, 3.70; N, 17.28. Found: C, 58.37; H, 3.98; N, 16.89%; IR: cm^{-1} 1386.1 [Ar-OH], 1687.5 [C=O], 3202.8 [Aromatic CH- Stretching], 3431.1 [NH-Stretching]; ^{1}HNMR: 89.364 [Py-C=O]; 7.02 [OH in Benzene ring]; 8.588 [O=C-NH-C=O].

N-Acetyal Pyrazinamide (3). Sodium Pyrazinamide and acetyl chloride were taken in equimolar proportion. i.e. 0.27 gm of acetyl per 0.5 gm of Sodium pyrazinamide. These two compounds were mixed and mixture was heated for a very short time. A light cream coloured compound formed separated on cooling which was then washed and recrystallized with alcohol to get fine powder. Yield 0.34 gm. Per 0.5 gm. of Sodium pyrozinamide, M.P. 160-163°C. Anal. Calcd. For C_7 H_7 N_3 O_2: C, 50.92; H, 4.24; N, 25.48. Found: C, 50.12; H, 3.92; N, 25.33%; IR: cm^{-1} 1430.6 [Banding vibration frequency for CH_3 group], 1675.8 [C=O]; 3162.8 [Aromatic CH-Stretching]; 3421.6 [NH-Stretching]; ^{1}HNMR: 89.267 [Py-C=O]; 8.705 [O=C-NH-C=O]; 3.331 [CH_3-C=O], Characterization data of these compounds are presented in table 1.

Table 1 : Characteristics and elemental analysis of PZA Derivatives

Compound	Yield gm./0.5gm. of Na-PZA	M.P. (^{0}C)	Mol. Formula	Found (%) [Calcd]		
				C	H	N
1	0.85	100-101	C_{12} H_9 N_3 O	62.6 [63.45]	3.68 [3.96]	18.63 [18.49]
2	0.85	130-135	C_{12} H_9 N_3 O_3	58.37 [59.27]	3.98 [3.70]	16.89 [17.28]
3	0.34	160-163	C_7 H_7 N_3 O_2	50.12 [50.92]	3.92 [4.24]	25.33 [25.48]

Table 2 : Antibacterial activity data of PZA Derivatives:

Invitro sensitivity of Derivatives of PZA on L-J medium [Drug containing L-J]

Compound	Suspension	Observation Days					
		7	14	21	28	35	42
INH	S_1	-	+	++	++±	+++	++++
	S_2	-	+	++	++±	+++	++++
	S_3	-	+	+	+±	++	+++
	S_4	-	+	±	+	+	++
	S_5	-	-	-	-	+	+
1	S_1	NG	NG	NG	NG	NG	NG
	S_2	-	-	-	-	-	-
	S_3	-	-	-	-	-	-
	S_4	-	-	-	-	-	-
	S_5	-	-	-	-	-	-
2	S_1	-	+	++	++	+++	+++
	S_2	-	+	+	++	++	+++
	S_3	-	-	-	+	+	+
	S_4	-	-	-	-	-	-
	S_5	-	-	-	-	-	-
3	S_1	-	+	+	++	++	++
	S_2	-	-	+	+	++	++
	S_3	-	-	-	+	+	+
	S_4	-	-	-	-	-	-
	S_5	-	-	-	-	-	-

Growth was recorded as +++ confluent growth

++ more then 100 colonies

1-99 the actual number of colonies

PZA [Control, Drug fee, L-J]

RESULT AND DISCUSSION

All the Synthesized derivatives of PZA were screened for their antibacterial behaviour against mycobacterium tuberculosis. "Proportion method" was used for the antibacterial activity. The method used to study the sensitivity was based on Lowenstein-Jensen medium[10,11]. The observations regarding the sensitivity of the compounds are based on the growth of the bacteria in different suspensions prepared by adding varying concentration of the compound and the days to inhibit the growth. Five suspensions (S_1,S_2,S_3,S_4,S_5, as indicated in table 2) were used for all proportion sensitivity tests (Where 4mg/ml of moist bacteria is present in the first suspension S, and the ratio of concentration in

others suspension as compared to S_1, is 1: 10:100:1000:10000 in terms of dilutions.) Standard antibacterial PZA was also tested under similar conditions for comparison. On the basis of the observations as shown in Table 2, synthesized derivative 1 (N-Benzoyl PZA) was found more active against bacteria in comparison to compound 2 (N-Salicyl PZA) and 3 (N-Acetyl PZA) as it inhibit the growth at each [every] concentration of the derivative] all levels. Compound 2 (N-S PZA) & compound 3 [N-A PZA] are least effective as compared to compound 1(N-B PZA). The two concentrations S4 and S5 compound 2 [N-S PZA] and compound 3 [N-A PZA] are found capable to inhibit the growth of bacteria even after six weeks.

ANALYSIS OF RESULTS :

PZA with Cinnamaldehyde, Furfuraldehyde and Acetophenone were synthesized which were characterized by elemental analysis and spectral studies. This organism can develop resistance to certain drugs, therefore the work is still continued to develop a safe drug which may prove more potent as compared to the previous drugs in this field. It is also noticed that susceptibility of mycobacterium tuberculosis against PZA and its derivatives are much useful in drug industry. On the basis of the observations it is found that bacteria in comparison to certain compounds under study as it inhibit the growth for the longest period for three weeks only, where the compounds like Isonicotinyl and Cinnamylidene Hydrazone inhibit the growth up to four weeks at same level of concentration.

CONCLUSION:

From above study it was concluded that the antibacterial potential of other synthesized compound 2 (N-Salicyl PZA) and compound 3 (N-Acetyl PZA) were not found capable to inhibit the growth of bacteria completely except. Compound 1 [N-Benzoyl PZA]. Compound 2 [N-Salicyl PZA] and compound 3 [N-Acetyl PZA] are not active enough as required for the complete inhibition of the growth of bacteria at lower concentrations. But control the growth of bacteria in higher concentration only the higher upto five weeks. This work is in agreement with the work of other coworks [13]. Certain more applications can be recorded using more potential values. This work leads to new frontier in the drug Industry and Biochemistry etc.

REFERENCES:

[1]. A. Schatz, E. Bugie, S.A. Waksman, Streptomekin, A Substance exhibiting antibiotic activity against Gram Positive and Gram negative bacteria, J.Proc. Soc. Exptl. Biol. Med. **1944**, 55-56.

[2]. H.H. Fox, JT Gibas, Synthetic tuberculostats V. Alkylidene derivatives of isonicoting hydrazinc, J.Org. Chem. **1953**, 17, 1653.

[3]. S. Kushner, H. Dalalain, J.L. Sanjurjo, F.L. Bach, Jr. S.R. Sarif, V.K. Smith, Jr. and J.H. Williams, J. Am. Chem. Soc., **1952**, 74, 3617.

[4]. L. Malone, A. Schorr, H. Lindh, D. Mckinzie, J.S. Kiser and J.H. Williams, Am. Rev. Tuber, **1952**, 65, 511.

[5]. R.L. Yeager, W.G.C. Munroe and F.l. Dissau, Am. Rev. Tuber, **1952**, 65, 523.

[6]. R.M. McCune, Jr. and Tompsett, J. Exptl. Med. **1956**, 104, 737-763.

[7]. J. Bernstein, W.A. Lott, B.A. Steinberg and H.L. Yale, Effect of isoniazid on tuberculosis lesions of the Kidneys, J. Am. Rev. Tuberc. **1952**, 65, 357.

[8]. P. Sensi, N. Maggi, S. Furesz and G. Mffi, Antimicrob, agents chemotherapy, **1967**, 699, 1966.

[9]. J.P. Thomas, C.O. Baughn, R.G. Wilkinson and R.g. Shepherd, A new synthetic compound with antituberculous activity in mica, J.Am Rev Res pir Dis, **1961**, 83,899.

[10]. S.P. Tripathy, Mithcison, D.A. Mai, N.G.K. Radhakrishna and S. Subbamml, A comparison of various methods of sensitivity of M. Tuberculosis to Isoniazid. J. Tubercle, **1970**, 51, 375.

[11]. Thelerno diallo, Menonli Adjobimey, Rovina Reeslami, Anete Trajman, Safety and side effects of rifaripin versus isonizid, N. Engl. J. Med. **2018**, 379 (5), 454-463.

[12] Timothy R. Sterling, M. Elsa Villarino, Andrey S. Borisov, three months of refapantive and isoniazid for lalent-tuber-culosis inifactum, N. Engl. J. Med. **2011**, (23), 2155-2166.

[13]. K.P. Tiwari, Dielectric Parameters and Intermolecular Interactions in Binary Mixture of Hydroxyl Groups in Polar Liquids, Jorn. of Appl. Chem., **2016,** 5 (6): 1302-1306.