**Title: Synthesis and evaluation of antibacterial activity of new pyrazolo[3,4-*b*]pyridine and pyrazolo[3,4-*d*]pyrimidine derivatives**

**Author:** Hamid Beyzaei,a Reza Aryan,a Mohammadreza Moghaddam-Manesh,b Behzad Ghasemic

a *Department of Chemistry, Faculty of Science, University of Zabol, Zabol, Iran*

b *Young Researchers and Elite Club, Kerman Branch, Islamic Azad University, Kerman, Iran*

c *Young Researchers and Elite Club, Neyshabur Branch, Islamic Azad University, Neyshabur, Iran*

**E-mail:** hbeyzaei@yahoo.com

**Date:** June 2017

**Introduction**

Presence of pyrazolo[3,4-*b*]pyridines and pyrazolo[3,4-*d*]pyrimidines in a variety of biologically active compounds such as anticancer, antimicrobial, antiparasitic, anti-inflammatory and antidiabetic agents, have attracted the attention of many synthetic organic chemists and medical researchers. Various synthetic strategies were developed for their preparation, but more methods are demand. In this project, two efficient methods to prepare some novel 6-substituted 3-methyl-1-phenyl-1*H*-pyrazolo[3,4-*b*]pyridin-4-amines and 6-substituted 4-imino-3-methyl-1-phenyl-1,4-dihydro-5*H*-pyrazolo[3,4-*d*]pyrimidin-5-amines has been proposed *via* cyclocondensation of 5-amino-3-methyl-1-phenyl-1*H*-pyrazole-4-carbonitrile with various methyl ketones and hydrazides in ethanolic sodium ethoxide. Inhibitory activity of the synthesized heterocyclic products was evaluated against a broad variety of pathogenic bacteria.





**Methods**

5-Amino-pyrazole-4-carbonitrile as starting material was prepared in one-pot procedure from the reaction of malononitrile, triethyl orthoacetate, phenylhydrazine and a few drops of acetic acid in ethanolic solution after refluxing for 12 h. The cyclocondensation of this pyrazole with hydrazides or methyl ketones in 0.5 M NaOEt/EtOH medium produced respectively pyrazolo[3,4-d]pyrimidines and pyrazolo[3,4-b]pyridines in moderate yields.

The antibacterial activities of heterocycles were determined by employing broth microdilution and disk diffusion methods, according to CLSI (Clinical and Laboratory Standards Institute) guidelines M07-A9, M26-A and M02-A11. The stock solutions of all derivatives and antibiotics were respectively prepared in 10% DMSO and double-distilled water at concentrations of 9011 and 17.6 μg/mL.

**Results**

In this study, a new and efficient two step procedure was proposed for the synthesis of some novel pyrazolo[3,4-b]pyridine and pyrazolo[3,4-d]pyrimidine derivatives. The chemical structure of all compounds was deduced from 1H NMR, 13C NMR, FT-IR and elemental analyses. The in vitro antibacterial activities of the newly synthesized derivatives were evaluated against a wide range of Gram-positive and -negative bacterial strains of various genera, which then compared with current antibiotics and reported as inhibition zone diameter (IZD), minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) values. The derivatives showed moderate to good inhibitory activities against pathogenic bacteria.