Preparation and Characterization of New Formazan Compounds Derivatives from 4-Methyl-O-Phenylenediamine and Study of Their Biological Activity

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Abstract
This study involves the synthesis of some formazan derivatives starting from reacting 4-Methyl-o-phenylenediamine with anthranilic acid to gate imidazole derivative(Ma), (Ma) react with chloro acetyl chloride to give compound (Mb) also (Mb) react with hydrazine hydrate to give compound (Mc) then (Mc) react with 4-Bromo benzaldehyde to product Schiff base derivative (M), then (M) react with different amine compounds to get formazan derivatives . All these compounds are characterized by, Fourier transforms infrared spectroscopy (FTIR), 1HMNR. After that , we study the biological activity for all formazan derivatives to ward two different kinds of bacteria.
Keywords: Schiff base, Formazan, Azo compounds ,bacteria

1. Introduction

Formazans are an important type of organic compound and they contain a distinctive chain of atoms,having th e general formula (-N=N-C=N-N-), (1),separately Bamberger and Von Pechman synthesized the first formazan afterward there have been many formazans synthesized and inspected(2), Formazan and their derived metal compound are dyed compounds. Color vision is attributed to π-π* transitions of π electrons in the formazan skeleton ,Formazans and their metal compound are ranging in color from red to orange as well as blue color(3). Schiff bases given name by Hugo Schiff 1834–1915, the German chemist who synthesized the first organs of this class of products containing an (azomethine) or (imine) Schiff involved the condensation of(a carbony l compound) aldehyde or ketone with a primary amine under azeotropic distillation (4), Schiff bases have premium characteristics, structural similarities with natural biological substances relatively and simple preparation procedures (5). Azo compounds are described by the presence of the azo group (–N=N–) in their structure, couple with two, special or identical mono- or polycyclic aromatic or heteroaromatic systems. So that of their specific physicochemical flavor and biologica l activities, they have got the wide application in pharmaceutical, cosmetic, painter or texture industry and analytical chemistry. but, the most typical and popular field of utility remains as their coloring function. The medical value of azo compounds is well famed for their antibiotic , antifungal , and anti-HIV properties(6), Azo dyes are the widely used dyes and counting for more than 60 % of total dyes(7), Color of azo dyes from to chromophores , active groups that absorb light giving bright colors, especially red, orange and yellow(8)[1].

2. 2. Materials

" (FTIR) Spectra (400 -4000 cm^{-1}) in KBr disk were recorded on SHIMADZU FTIR-8400S Fourier transform. ^{13}C-NMR and ^{1}HNMR were recorded on Varian Agilent USA at (500MHz) with (DMSO-d6) measurements were made at Department of Chemistry, Tehran University, Iran."

2.1 Preparation of compound(Ma)(9)

Mix equal moles of 4-Methyl-o-phenylene diamine (0.0818 mol,1gm) with anthranilic acid (0.0818 mol,1.225 gm) in 20 ml of 4N.HCl with reflex for 1 hours where the reaction process was followed using TLC technique , The precipitation was then filtered and recrystallization by using methanol , to resultedlight brown precipitation and molecular weight of 223.28 and a percentage of 90% were obtained,the melting point was with in rang 180-183°C and R_f = 0.34.

2.2.Preparation of compound (Mb) (10),(11)

Mix equal moles of chloroacetylchloride compounds (0.0045 mol ,0.37 ml) with compound (M_a) in 20 ml of dry benzene at room temperature with stirring for 3hours The reaction was followed by TLC technique.The precipitation was then filtered and recrystallization by with absolute ethanol to resulte ddark brown precipitation and molecular weight 299.76 and aprecentage 87% were obtained the melting point was with in range 225-228 °C and R_f = 0.43.

2.3.Preparation of compound (Mc) (12),(13)

Mix equal moles of hydrazine hydrate (0.0034 mol ,0.105 ml) with compound M_b (0.0034,1gm) at room temperature for (9)hours wherethe reaction was followed by TLC technique.The precipitation was then filtered and recrystallization using $CHCl_3$.MeOH to resulted brown precipitation and a

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precentage 78% were obtained the melting point was with in range 190-193 °C and R_f= 0.73.

2.4.Preparation of Schiff base compound(M) (14),(15)

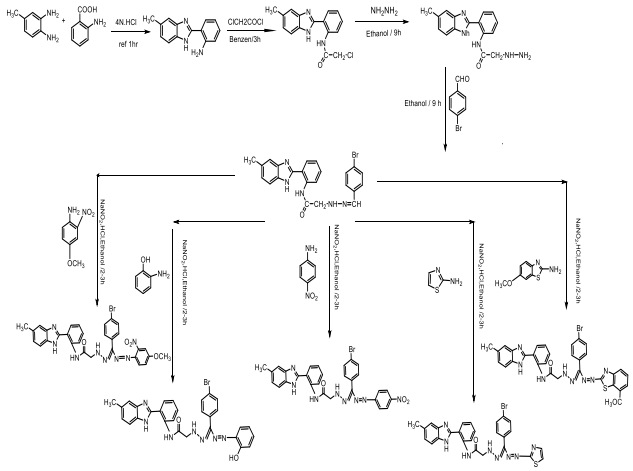
Mix equal moles of compound (M_c) (1gm , 0.003 mol) with 4-bromo benzaldehyde (0.56gm,0.003mol) and reflex at temperatureof 78 C^0 for 9hours where the reaction was followed by TLC technique.The precipitation was then filtered and recrystallization using ethanol absolute to resulted orange precipitation and molecular weight 463.4 and aprecentage 90% were obtained the melting point was with in range 324 -327 °C and R_f= 0.52.

2.5.Preparation of Dizonum salt (f) [16],[17]

Dissolve 0.002 mol of the aromatic amin (2-amino-6-methoxybenzothiazole,2-aminothiazole,2-aminophenol,4-nitroaniline,4-methoxy-2-nitroaniline) is dissolved in (5ml HCl+8ml) distilled water the solution is cooled to atempreature of (0-5° C) after that added to 0.002 mol sodium nitrate dissolved in 5ml distilled water drop by drop the mixture and leave for 20 minutes inside the refeigerator.

2.6.Preparation of Formazan Derivatives (18),(19)

Dissolve (0.002)mol of the Schiff base derivative(M) to the preparation of derivative from M_1-M_5 in 10 ml dioxane + 20 ml ethanol .The dizonium salt produced from the above step is added to the solution of the Schiff base drop by drop in snow bath 0-5 C^0 .It is left with stirring for aperiod of 2-3.5 hours (scheme(1)) , The reaction process was followed using TLC technique .Obtain color derivatives to be weighed and calculate the percentage and physical properties as show in Table I

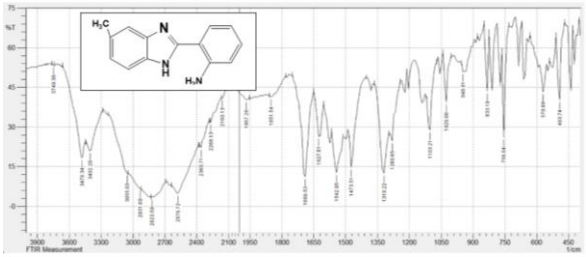


scheme(1):- prepare of some formazan derivatives

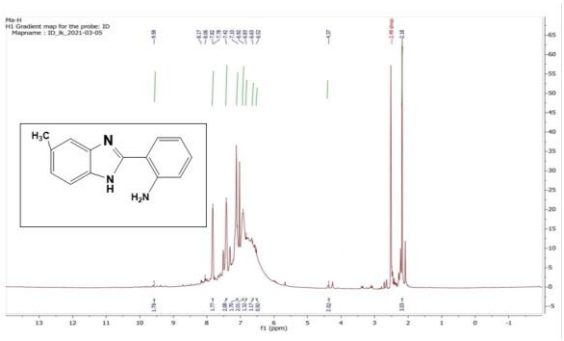
3. Results and Discussion

Characterization of the compound (Ma) 2-(5-methyl-1H-benzo[d]imidazol-2-yl)aniline

FT-IR spectrum date for derivative (Ma)show absorption packs at (3479- 3402 cm-1) for NH2 and N-H overlapping ,3055.03 cm-1 for (Ar-H) , 2931.60 cm-1 for (C-H) in CH3 , 1689.53 cm-1 for (C=N) , 1542.95 for (C=C) aromaticshow in fig(1) .1HMNR Spectrum data of compound (Ma) show 4.3ppm (S, 2H,(NH2),9.5ppm(S,1H,NH) for imidazole,2.1ppm (S,3H,CH3), 6.5 - 8.1 ppm(M,7H,Ar-H) show in fig(2).



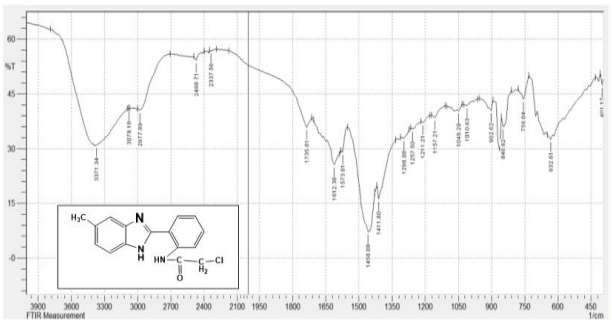
Fig(1):- FTIR spectrum of compound(Ma



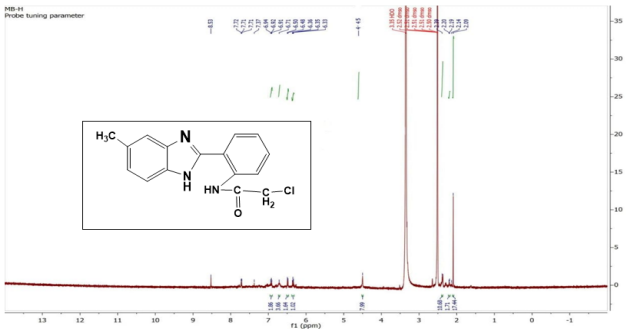
Fig(2):-(1 H-NMR) spectrum of compound(Ma)

Characterization of the compound (Mb) 2-chloro-N-(2-(5-methyl-1H-benzo[d]imidazol-2-yl)phenyl)acetamide

FT-IR spectrum date for derivative (Mb) show band at 3371.34cm-1 for(N-H) ,3085 cm-1 for (Ar-H) , 2977.89 cm-1 for (C-H) in CH3, 1735.81 for (C=O) , 1612.38 cm-1 for (C=N), 1573.81 cm-1 for (C=C),756.04 cm-1 for (C-Cl) show in fig(3) .1HMNR Spectrum data of compound (Mb) show 4.45ppm (S, 1H,(NH- C=O)) ,8.5ppm (S,1H,NH) for imidazole,2.1ppm (S,3H,CH3) ,2.3ppm (S,3H,CH2) show in fig(4).



Fig(3) :-FTIR spectrum of compound(Mb)

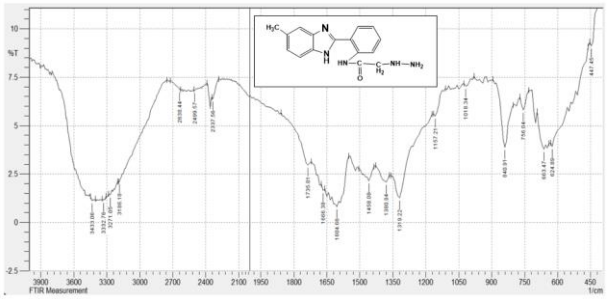


Fig(4):- (^{1} H-NMR) spectrum of compound(M_b)

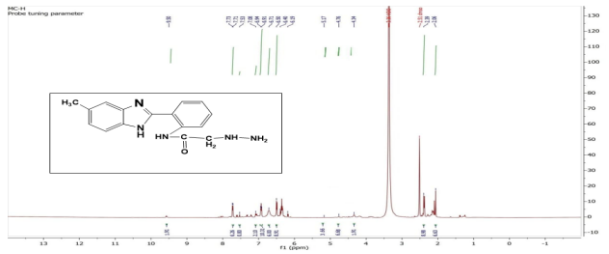
Characterization of compound (M_c

2-hydrazineyl-N-(2-(5-methyl-1H-benzo[d]imidazol-2-yl)phenyl)acetamide

FT-IR spectrum date for derivative (Mc) show absorption packs at (3433.06-3332.76cm-1) for(N-H)to the returning group NH2and N-H ,3078.47 cm-1 for (Ar-H) ,2938.44 cm-1 for (C-H) in CH3 ,1735.81 cm- 1 for (C=O) 1666.38 cm-1 for (C=N) ,1604.66 for (C=C) aromaticshow in fig(5) . 1HMNR Spectrum data of compound (Mc) show 2.06 ppm (S,3H,CH3), 9.5 ppm (S,1H,NH) for imidazole , 5.1ppm (S,2H,NH2),4.7ppm (S,1H,NH-Ar),4.3ppm (S,1H,NH-NH2),2.3ppm (S,2H,CH2) show in fig(6).



Fig(5):- FTIR spectrum of compound(M_c)

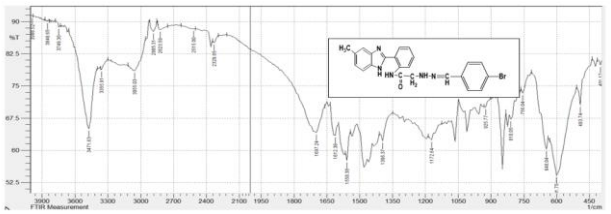


Fig(6):-(^{1} H-NMR) spectrum of compound(M_c)

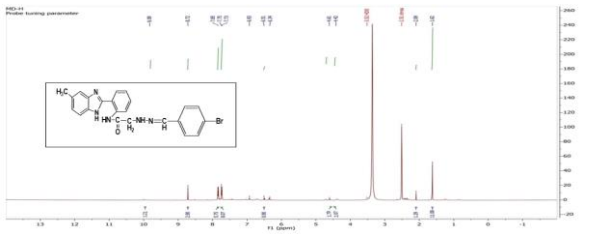
Characterization of compound (M)

2-(2-(4-chlorobenzylidene)hydrazineyl)-N-(2-(5-methyl-1H-benzo[d]imidazol-2-yl)phenyl)acetamide

FT-IR spectrum date for derivative (M) show band at (3471.63cm-1)for(N-H) ,3055.03 cm-1 for (Ar-H) ,2885.31 cm-1 for (C-H) in CH3 , 1697.24cm-1 for (C=O) , 1612. 38cm-1 for (C=N), 1558.38cm-1 for (C=C) aromatic,648.07 cm-1 for (C-Br) show in fig(7) . 1HMNR Spectrum data of compound (M) show 9.09 ppm (S, 1H, N-H)for imidazole)),4.6ppm (S,1H,NH-C=O),4.2ppm (S,1H,NH-N), 6.3 - 8.7 ppm (M,11H,Ar-H),1.6pmpp(S,3H,CH3) show in fig(8).



Fig(7) :-FTIR spectrum of compound(M)

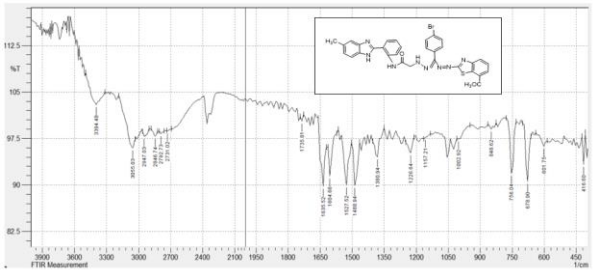


Fig(8):-(1 H-NMR) spectrum of compound(M)

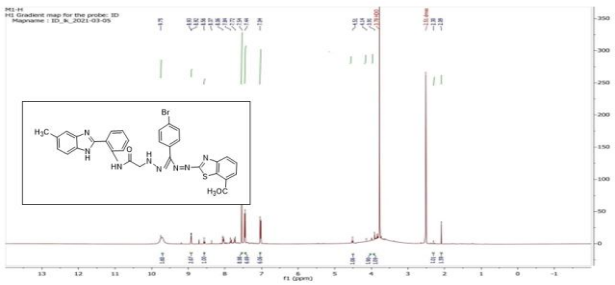
Characterization of compound (M_1)

)E)-1-(7-((l6-oxidaneylidyne)methyl)benzo[d]thiazol-2-yl)-3-(4-bromophenyl)-5-(2-((2-(5-methyl-1H-benzo[d]imidazol-2-yl)phenyl)amino)-2-oxoethyl)formazan

FT-IR spectrum date for derivative (M1) show band at (3394.48cm-1) for(N-H) ,3055.03 cm-1 for (Ar-H) ,2947.03 cm-1 for (C-H) in CH3,1735.81 cm-1 for (C=O) ,1635.52 cm-1 for (C=N) ,1527.52 for (C=C) ,1488.94 cm-1 for(N=N))),1226.64cm-1 for (C-S) ,601.75 cm-1 for (C-Br) show in fig(9) . 1HMNR Spectrum data of compound (M1) show 9.7 ppm (S, 1H,(N-H for imidazole)),4.5ppm(S,1H,NH-C=O),4.1ppm ,(S,1H,NH-N), 7- 8.9 ppm,(M,14H,Ar-H),2.3ppm, (S,3H,CH3),2.09ppm, (S,2H,CH2),3.9ppm,(S,3H,OCH3) show in fig(10).



Fig(9):- FTIR spectrum of compound(M_1)

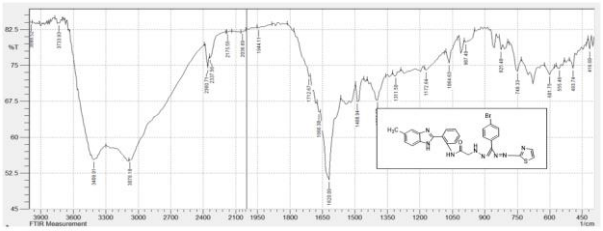


Fig(10):-(1 H-NMR) spectrum of compound(M1)

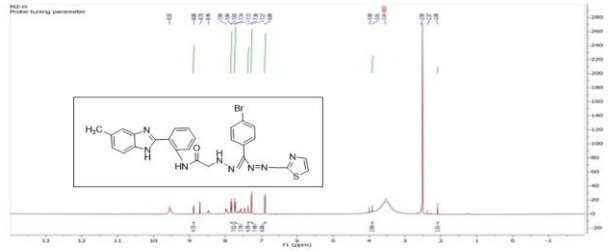
Characterization of compound (M_2)

)E)-3-(4-bromophenyl)-5-(2-((2-(5-methyl-1H-benzo[d]imidazol-2-yl)phenyl)amino)-2-oxoethyl)-1-(thiazol-2-yl)formazan

FT-IR spectrum date for derivative (M_2) show band at (3409.91cm^{-1}) for(N-H) ,3078.18cm^{-1} for (Ar-H) ,2907 cm^{-1} for (C-H) in CH_3,1712.67 cm^{-1} for (C=O) , 1666.38cm^{-1} for (C=N) ,1620.09 for (C=C) ,1488.94 cm^{-1} for(N=N)),1272.93 cm^{-1} for (C-S),601.75 cm^{-1} for (C-Br) show in fig(11). ^{1}HMNR Spectrum data of compound (M_2) show 9.5 ppm (S, 1H,(N-H for imidazole)),3.99ppm(S,1H,NH-C=O),3.91ppm ,(S,1H,NH-N), 6.8- 8.7 ppm,(M,1H,Ar-H),2.09ppm, (S,2H,CH_2),2. 3ppm, (S,3H,CH_3) show in fig(12)[2] .



Fig(11):_ FTIR spectrum of compound(M_2)

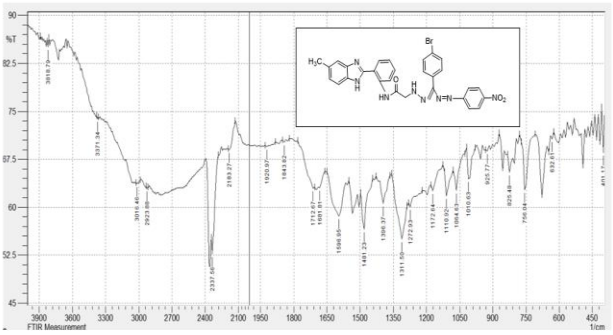


Fig(12):-(¹ H-NMR) spectrum of compound(M_2)

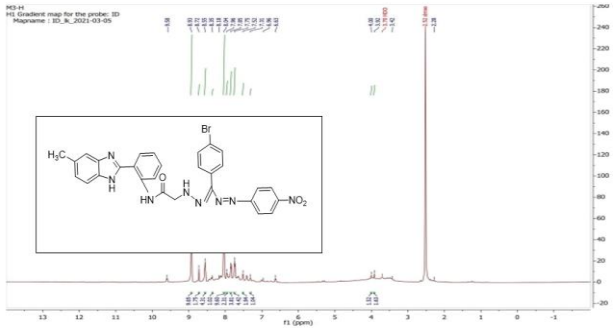
Characterization of compound (M_3)

)E)-3-(4-bromophenyl)-5-(2-((2-(5-methyl-1H-benzo[d]imidazol-2-yl)phenyl)amino)-2-oxoethyl)-1-(4-nitrophenyl)formazan

FT-IR spectrum date for derivative (M_3) show band at (3371.34cm^{-1}) for (N-H) ,3016.46 cm^{-1} for (Ar-H) ,2932.88 cm^{-1} for (C-H) in CH_3 ,1712.67 cm^{-1} for (C=O) ,1681.81 cm^{-1} for (C=N) ,1596.95 for (C=C) , 1481.23cm^{-1} for(N=N)),632.61cm^{-1} for (C-Br) show in fig(13) . ^{1}HMNR Spectrum data of compound (M_3) show 9.5 ppm (S, 1H,(N-H for imidazole)),4ppm(S,1H,NH-C=O),3.9ppm ,(S,1H,NH-N), 6.6-8.9 ppm, (M,15H,Ar-H),2.2ppm, (S,3H,CH_3),3.4ppm, (S,2H,CH_2) show in fig(14).



Fig(13):- FTIR spectrum of compound(M_3)

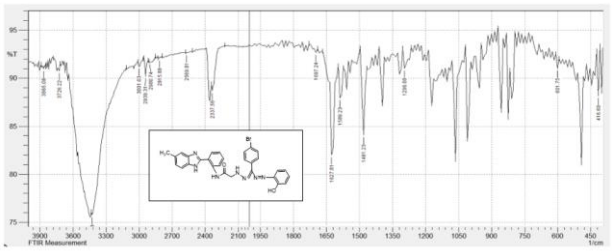


Fig(14):-(¹ H-NMR) spectrum of compound(M_3)

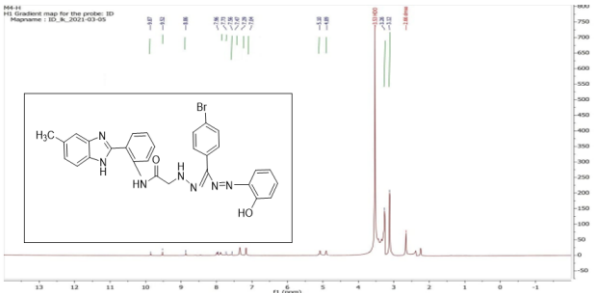
Characterization of compound (M_4)

)E)-3-(4-bromophenyl)-1-(2-hydroxyphenyl)-5-(2-((2-(5-methyl-1H-benzo[d]imidazol-2-yl)phenyl)amino)-2-oxoethyl)formazan

FT-IR spectrum date for derivative (M_4) show band at (3425.34cm^{-1}) for(N-H), 3001.03 cm^{-1} for (Ar-H) , 2939.31 cm^{-1} for (C-H) in CH_3 ,1697.24 cm^{-1} for (C=O) ,1627.81 cm^{-1} for (C=N) ,1589.23 for (C=C) ,1481.23 cm^{-1} for(N=N),),601.75 cm^{-1} for (C-Br) show in fig(15). ^{1}HMNR Spectrum data o f compound (M_4) show 9.7 ppm (S, 1H,(N-H for imidazole)), 9.8ppm,S,1H,OH, ,5.1ppm(S,1H,NH-C=O),4.8ppm ,(S,1H,NH-N), 7- 7.9 ppm,(M,15H,Ar-H),3.1ppm, (S,3H,CH_3),3.2ppm, (S,2H,CH_2) show in fig(16).



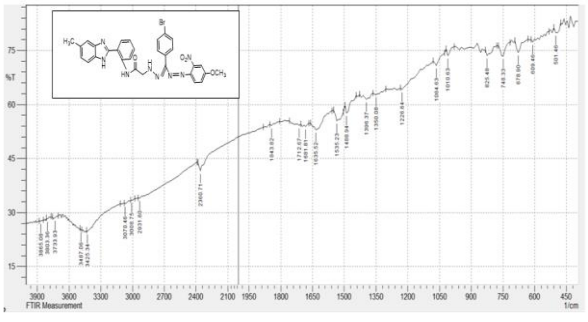
Fig(15) :-FTIR spectrum of compound(M_4)



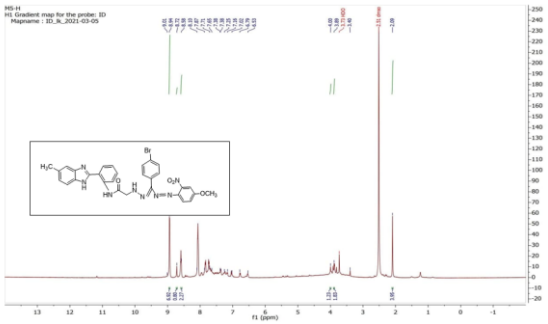
Fig(16):-(¹ H-NMR) spectrum of compound(M_4)

Characterization of compound (M5) (E)-3-(4-bromophenyl)-1-(4-methoxy-2-nitrophenyl)-5-(2-((2-(5-methyl-1H-benzo[d]imidazol-2-yl)phenyl)amino)-2-oxoethyl)formazan

FT-IR spectrum date for derivative (M5) show band at (3487.06cm-1) for (N-H) , 3008.75cm-1 for (Ar-H) , 2931.60cm-1 for (C-H) in CH3 , 1712.67cm-1 for (C=O) , 1681.81cm-1 for (C=N) , 1635.52cm-1 for (C=C) , 1488.94cm-1 for(N=N), (1535.23-1350.08 cm-1) for (R-NO2),),609.46 cm-1 for (C-Br) show in fig(17) . 1HMNR Spectrum data of compound (M5) show 9.01 ppm (S, 1H,(N-H for imidazole)), 4ppm (S,1H,NH- C=O),3.89ppm ,(S,1H,NH-N), ppm,(M,14H,Ar-H),1.23ppm, (S,3H,CH3) , 2.09ppm, (S,2H,CH2),3.4ppm,(S,3H,OCH3) show in fig(18)[3].



Fig(17):- FTIR spectrum of compound(M5)



Fig(18):-(¹ H-NMR) spectrum of compound(M_5)

4. Biological activity

4.1 :-Antibacterial

the results of the antibacterial activity are shown in the fig (19) Table (1) the results of the antibacterial activity for (1-5) derivatives				
Mm	Staph aureus	Mm	E.Coli	Comp.NO
13	++	16	++	1
14	++	15	++	2
13	++	15	++	3
11	++	15	++	4
12	++	15	++	5
32	+++	40	+++	Penicillin
"+= (5-10)mm =slightly active, ++= (11-20)mm moderately, +++ = More than 20 , good active"				

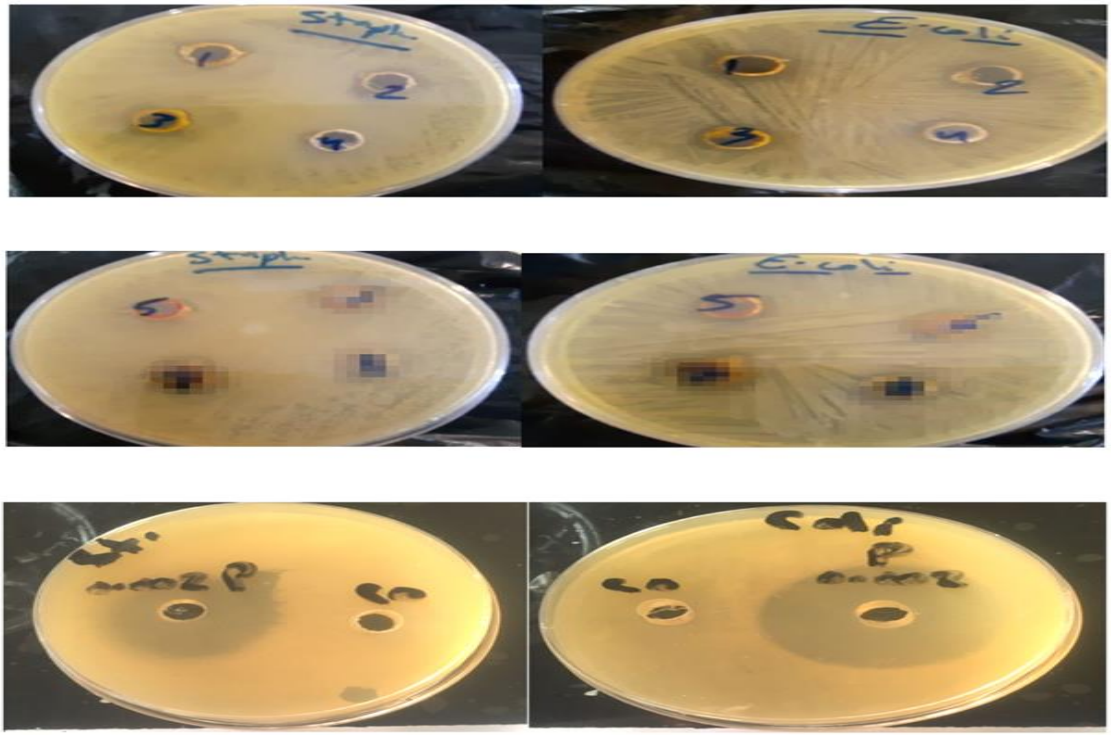


Fig (19) effect compounds(E.Coli) aginst and (Staph aurous) aginst

Table (2) Prorates of derivtive (1-7)derivetives						
Yield %	Color	R_f	M.p (^{0}C)	M.W	M.F	Comp.
78	Brown	0.14	110	2 682.6	C33H30BrN8O2S	1
62	Dark Brown	0.26	241	602.11	C28H26BrN8OS	2
61	Brown	0.27	197	625.18	C32H32BrN7O2	3
70	Light Brown	0.22	104	655.56	C32 H31BrN8O3	4
75	Brown reddish	0.15	97	684.58	C33H32BrN8O4	5

5. Conclusion

1 -Formazan compounds areb vere stable
2 The prepared compounds have a high biological activityas antibacterial.
3 -The derivatives have a distinctive color because they contain azo and formazan group

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