Synthesis and Antibacterial Activity Evaluation of Several New Sulfonamido Naphthalimides Linked to Benzothiazole or Oxadiazole

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ABSTRACT

In this work several new sulfonamido naphthalimides linked to benzothiazole or oxadiazole cycles were synthesized via multistep synthesis. The first step involved synthesis of compound [1] N-phenylnaphthalimide via reaction of aniline with naphthalic anhydride at high temperature in the presence of glacial acetic acid. In the second step compound [1] was introduced in reaction with chlorosulfonic acid producing compound [2] 4-(N-naphthalimidyl) phenyl sulfonyl chloride which inturn introduced subsequently in the third step in reaction with 2-amino-5-substituted-1,3,4-oxadiazoles producing the target naphthalimides [10, 11] or with substituted 2-aminobenzothiazoles producing the target naphthalimides [12-16]. Results of antibacterial activity study of the newly synthesized naphthalimides indicated that they posses high antibacterial activity.

تحضير وتعيين الفعالية المضادة للبكتريا لعدد من سلفون اميدونفثال ايمايدات الجديدة المرتبطة بمكونة البنزوثايازول او الاوكسادايازول

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الكلمات المفتاحية: نفثال ايمايدات، 1،3،4-اوكسادايازول، بنزوثايازول

الخلاصة

تم في هذا البحث تحضير سلفون اميدو نفثال ايمايدات جديدة مرتبطة بمكونة الاوكسادايازول او البنزوثايازول بوساطة التحضير المتعدد الخطوات حيث تم في الخطوة الاولى تحضير المركب [1] N-فنيل نفثال ايمايد وذلك من تفاعل الانيلين مع انهيدريد النفثاليك في درجات حرارة عالية بوجود حامض الخليك الثلجي. في الخطوة الثانية تم ادخال مركب [1] في تفاعل مع كلوروحامض السلفونيك مما اسفر عن تكوين مركب [2] 4-(N-نفثال ايمديل) فنيل كلوريد السلفونيل والذي بدوره تم ادخاله لاحقا في تفاعل اما مع مركبات 2-امينو-5-معوض-1،3،4-اوكسادايازول للحصول على الايمايدات الهدف [10،11] او في تفاعل مع مركبات 2-امينوبنزوثايازول للحصول على الايمايدات الهدف [12-16]. اوضحت نتائج دراسة الفعالية المضادة للبكتريا للايمايدات المحضرة بانها ذات فعالية عالية ضد انواع البكتريا قيد الدراسة.

1- INTRODUCTION

Naphthalimides first discovered by Brana have shown anticancer activity against a variety of murine and human tumor cells (1,2). Besides many 1,8-naphthalimides and bisnaphthalimides have shown antibacterial, antifungal and analgesic properties (3,4).

On the other hand 1,3,4-oxadiazoles belong to a group of heterocycles that have been attracting attention for the last two decades due to their wide range of biological interactions (5,6). Many of them exhibit antibacterial (7), anticonvulsant (8), anticancer activities and are used to fight infections (9) involving AIDS.

Benzothiazoles also comprise a class of organic compounds with multiple applications and are of particular interest since great variety of these compounds display various biological activities

(10-13). Moreover sulfonamido group is very important to enhance some biological activities like antimalarial and antimicrobial (14).

Encouraged by all these observations it was though worthwhile to synthesize new compounds via incorporating naphthalimide and benzothiazole or oxadiazole nucleus linked together through sulfonamido group in the same framework and screened for their in vitro antibacterial activity.

2. MATERIALS AND METHODS

All chemicals used in this work were purchased from fluka and BDH and used without further purification.

Melting points were determined on Thomas Hoover apparatus and were uncorrected. FTIR spectra were recorded on SHIMADZU FTIR-8400 infrared spectrophotometer. ^{1}HNMR and ^{13}CNMR spectra are recorded on Bruker 300MHz instrument using tetramethylsilane (TMS) as the internal standard and DMSO-d_6 as solvent.

2.1. Preparation of N-phenyl-1,8-Naphthalimide [1]

A mixture of 1,8-naphthalic anhydride (0.01 mol, 1.98 g) and aniline (0.01 mol, 0.93 g) in glacial acetic acid (50 mL) was refluxed for 8 hrs (15). The resulted mixture was poured into cold water with stirring then the formed precipitate was filtered, washed with water, dried and finally recrystallized from ethanol. Yield (95%), m.p. (102-104)°C.

2.2. Preparation of 4-(N-Naphthalimidyl)phenyl sulfonyl chloride [2]

The titled compound was prepared according to literature (16) with some modifications. Chlorosulfonic acid (5 mL) was placed in a suitable round bottomed flask equipped with thermometer and dropping funnel containing (0.1 mol, 2.73 g) of N-phenyl naphthalimide dissolved in (30 mL) of chloroform. Flask content was cooled to (zero)°C then imide solution was added dropwise at such a rate that temperature of the well stirred mixture did not rise above (5)°C.

The reaction mixture was stirred for additional four hours then was allowed to stand overnight in the refrigerator. The resulted mixture was poured into crushed ice with stirring and the obtained oily layer was separated by extraction with chloroform then the solvent was evaporated and the formed precipitate was purified by recrystallization from petroleum ether (b.p. 60-80)°C. Yield (80%), (220°C decomposition).

2.3. Preparation of 5-substitited-2-amino-1,3,4-oxadiazoles [3,4]

The titled compounds were prepared according to literatures with some modifications (17) via two step synthesis. The first step involved synthesis of semicarbazones by refluxing a mixture of (0.01 mol, 1.11 g) of semicarbazide hydrochloride, (0.01 mol, 0.82 g) of sodium acetate and (0.01 mol) of aromatic aldehyde in (25 mL) of ethanol with stirring. The obtained precipitate was filtered, washed with ethanol and dried. In the second step bromine solution [(0.01 mol) of bromine dissolved in (8 mL) acetic acid] was added dropwise to a mixture of the prepared semicarbazone (0.01 mol) and sodium acetate (0.01 mol, 0.82 g) dissolved in (25 mL) of acetic acid with stirring then stirring was continued for additional two hours. The resulted solid was filtered, dired then recrystallized from a suitable solvent.

2.4. Preparation of substituted-2-aminobenzothiazoles [5-9]

Bromine solution [(0.025 mol) of bromine dissolved in (15 mL) acetic acid] was added dropwise to a mixture of (0.01 mol) of 4-substituted aniline, (0.01 mol, 0.76 g) of ammonium thiocyanate dissolved in (20 mL) of glacial acetic acid with stirring and cooling for two hours (18). The resulted mixture was diluted with distilled water then 10% NaOH solution was added with stirring until precipitation of the product. The formed precipitate was filtered, dried then recrystallized from a suitable solvent.

2.5. Preparation of N-[4-(5-substituted-1,3,4-oxadiazole-2-yl-sulfonamido)phenyl]-1,8-naphthalimides [10,11]

4-(N-naphthalimidyl)phenyl sulfonyl chloride (0.01 mol, 3.71 g) was added in portions to (0.01 mol) of 5-substituted-2-amino-1,3,4-oxadiazole dissolved in (30 mL) of pyridine with stirring and keeping temperature below (40°C). The resulted mixture was refluxed for three hours then was poured into excess cold water with stirring and the obtained precipitate was filtered, washed with water, dried and recystallized from a suitable solvent (16). Physical properties of compounds [10, 11] are listed in Table (1).

2.6. Preparation of N-[4-(substituted benzothiazole-2-yl-sulfonamido)phenyl]-1,8-naphthalimides [12-16]

The titled compounds were prepared by following the same procedure used in the preparation of compounds [10,11] except using of substituted-2-aminobenzothiazoles instead of 2-amino-1,3,4-oxadiazoles. The prepared imides were purified by recrystallization from a suitable solvent (16).

Physical properties of compounds [12-16] are listed in Table (1).

2.7. Antibacterial Activity Study

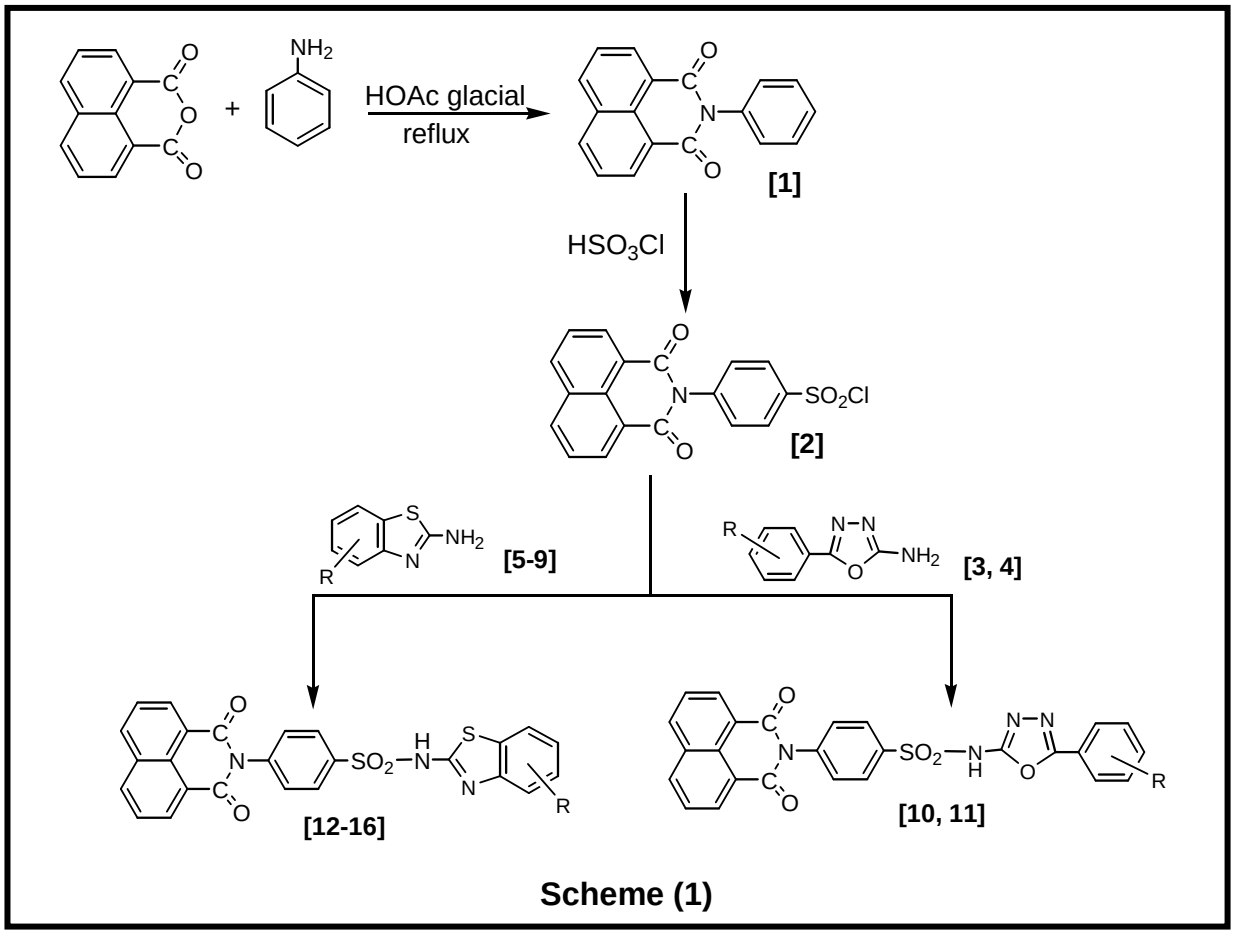
Antibacterial activities of the prepared naphthalimides [10-16] were determined against four different strains of bacteria including (*Staphylococcus aureus, Streptococcus pyogenes, Escherichia coli* and *Pseudomonas aureginosa*) using Ampicillin as standard drug. The cup plate method was used, DMSO was used as sample solution, sample size of all compounds was fixed at (0.1 mL) and the used concentration for tested compounds was 100 μg/mL. Using a sterilized cork borer cups were scooped out of agar medium contained in a petridish which was previously inoculated with the bacteria. The test compound solution (0.1 mL) was added in the cup and the petridishes were subsequently incubated at 37°C for 48 hrs. Zones of inhibition produced by each compound were measured in (mm) and the results are listed in Table (3).

3. RESULTS AND DICUSSION

Since naphthalimides, benzothiazoles, oxadiazoles and sulfonamides are known biologically active compounds having wide spectrum of medicinal and chemotherapeutic applications the target of this work is to build new molecules containing these active moieties via synthesis of new 1,8-naphthalimides containing both sulfonamide group and hetero ring (benzothiazole or oxadiazole).

The strategy used for performing this target involved preparation of N-phenyl-1,8-naphthalimide which inturn was introduced in reaction with chlorosulphonic acid producing naphthalimidyl phenyl sulfonyl chloride which subsequently attacked amino group in benzothiazole or oxadiazole compounds producing the desired sulfonamido naphthalimides.

This linear pathway strategy can be summarized in Scheme (1).



Scheme (1)

Compound [1] N-phenyl-1,8-naphthalimide was prepared at the first step via reaction of aniline with 1,8-naphthalic anhydride in boiling glacial acetic acid (15). The reaction was performed by nucleophilic attack of aniline amino group on carbonyl group in naphthalic anhydride producing N-phenyl naphthal amic acid which was not separated and instead introduced directly under the influence of glacial acetic acid and heat in dehydration reaction producing compound [1].

FTIR spectrum of compound [1] showed clear absorption bands at (1701, 1662) cm^{-1} due to ν(C=O) imide, (1585, 1624) cm^{-1} due to ν(C=C) aromatic and 1238 cm^{-1} due to ν(C-N) imide (19).

^{1}HNMR spectrum of compound [1] showed signals at (δ=7.38-8.5) ppm belong to aromatic protons while ^{13}CNMR spectrum showed signals at (δ=123-136.5) ppm due to aromatic carbons and at (δ= 164.13) ppm due to (C=O) imide carbons.

In the second step compound [1] was introduced in reaction with chlorosulfonic acid producing compound [2] 4-(N-naphthalimidyl)phenyl sulfonyl chloride.

The reaction was proceed through electrophilic substitution reaction of electron-deficient sulfur atom in HSO_3Cl on para position of phenyl ring in naphthalimide followed by elimination of water molecule.

FTIR spectrum of compound [2] showed clear absorption bands at (1705, 1662) cm^{-1} due to ν(C=O) imide, (1585) cm^{-1} due to ν(C=C) aromatic, (1354) cm^{-1} due to ν(C-N) imide, (1377 and 1192) cm^{-1} due to ν(SO_2)asym. and ν(SO_2)sym. respectively.

^{1}HNMR spectrum of compound [2] showed signals at (δ=7.2-8.1) ppm belong to aromatic protons while ^{13}CNMR spectrum showed signals at (δ=120.3-143.1) ppm due to aromatic carbons and at (δ= 167.4) ppm belong to (C=O) imide carbons.

In the third step compound [2] was introduced in reaction with 2-amino-1,3,4-oxadiazoles [3,4] and 2-aminobenzothiazoles [5-9] producing the target sulfonamido naphthalimides [10-11] and [12-16] respectively.

2-amino-5-substituted-1,3,4-oxadiazoles [3,4] and substituted-2-aminobenzothiazoles [5-9] used in this work were prepared according to literature procedures and their physical properties and spectral data are fitted with those reported in literatures.

Reaction of compound [2] with 2-amino-1,3,4-oxadiazoles or 2-amino benzothiazoles was proceed via nucleophilic attack of amino group in oxadiazole compounds [3,4] or in benzothiazole compounds [5-9] on sulfur atom in compound [2] followed by elimination of HCl molecule. Physical properties of compounds [10-16] are listed in Table (1).

FTIR spectra of compounds [10, 11] showed absorption bands at (3300-3407) cm^{-1} due to ν(NH) amide, (1705-1710) and (1660-1665) cm^{-1} due to ν(C=O) imide, (1600-1612) cm^{-1} due to ν(C=N) and (1585-1590) cm^{-1} due to ν(C=C) aromatic.

Other absorption bands appeared at (1355-1360) cm^{-1}, (1375-1380) cm^{-1}, (1157-1160) cm^{-1} and (1185-1190) cm^{-1} due to ν(C-N) imide, ν(SO_2)asym. ν(SO_2)sym. and ν(C-O) oxadiazole respectively. FTIR spectral data of compounds [10,11] are shown in Table (2).

^{1}HNMR spectrum of compound [11] showed signals at (δ= 6.69-8.83) ppm belong to aromatic and (NH) protons, while ^{13}CNMR spectrum of the same compound showed signals at (δ=123-131) ppm, (δ=134.89-136.5) ppm and (δ=164.1) ppm belong to aromatic carbons, two (C=N) and (C=O) imide carbons respectively.

FTIR spectra of compounds [12-16] showed absorption bands at (3275-3464) cm^{-1} due to ν(NH) sulfonamide, (1701-1708) cm^{-1}, (1662-1666) cm^{-1} due to ν(C=O) imide, (1589-1635) cm^{-1} due to ν(C=N) and (1539-1589) cm^{-1} due to ν(C=C) aromatic.

Other absorption bands appeared at (1307-1355) cm^{-1} due to ν(C-N) imide, (1354-1377)cm^{-1} due to ν(SO_2)asym., (1186-1192) cm^{-1} due to ν(SO_2)sym. and (651-698) cm^{-1} due to ν(C-S) thiazole.

^{1}HNMR spectrum of compound [12] showed signals at (δ= 2.06) and (δ= 7.44-8.52) ppm belong to protons of two methyl group and (NH) and aromatic protons while ^{13}CNMR spectrum showed signals at (δ= 15.28 and 18.0) ppm belong to two methyl groups carbons and signals at (δ= 112.2-137.7), (139.6) and (163.7) ppm belong to aromatic carbons, (C=N) and (C=O) imide carbons respectively.

^{1}HNMR spectrum of compound [13] showed signals at (δ= 7.3-8.5) and (11.7) ppm belong to aromatic protons and (NH) proton while ^{13}CNMR spectrum showed signals at (δ= 120.2-149) (164.6) and (166.8) ppm belong to aromatic carbons, (C=N) and (C=O) imide carbons respectively.

^{1}HNMR spectrum of compound [14] showed signals at (δ= 2.26) and (δ= 7.31-8.52) ppm belong to CH_3 protons and aromatic protons respectively while ^{13}CNMR spectrum showed signals at (δ= 21.51), (119.52-156.3), (161.4) and (163.83) ppm which belong to CH_3 carbon, aromatic carbons, (C=N) and (C=O) imide carbons respectively (19).

3- BIOLOGICAL STUDY

The cup plate method was employed in studying the antibacterial activity of the prepared imides against four strains of bacteria. Inhibition zone caused by each compound was measured in (mm) and the results are listed in Table (3).

The results indicated that compounds [12] and [14] showed very high activity against *S. aureus,* compounds [10] and [13] showed high activity against *S. aureus* also while compounds [10], [12], [14] and [16] showed high activity against *S. pyogenes.* Compound [11], [12] and [14] showed high activity against *P. aeruginosa* while compound [11] and [13] showed high activity against *E. coli.*

Table 1: Physical properties of compounds [10-16]

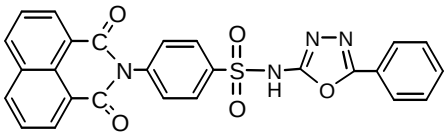
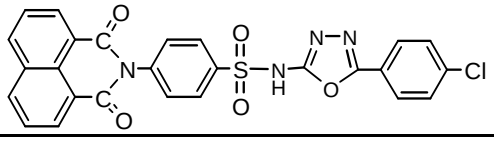
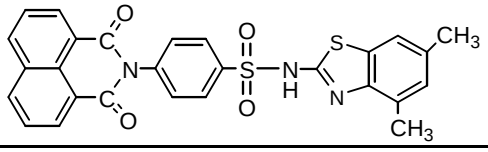
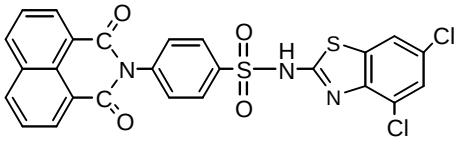
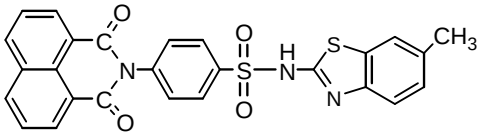
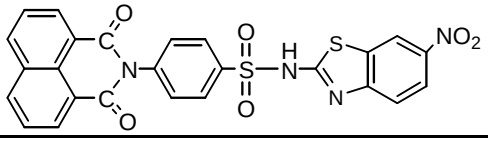
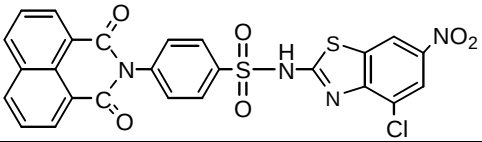
Comp. No.	Compound structure	Color	Melting point °C	Yield %	Solvent of recrystallization
10		Off white	194-196	75	Ethanol
11		Light brown	178-180	63	Ethanol
12		Dark brown	165-167	82	Ethanol
13		Brown	210-212	78	Chloroform
14		Light brown	191-193	80	Ethanol
15		Yellow	184-186	65	Ethanol
16		Greenish yellow	214-215	60	Chloroform

Table 2: FTIR spectral data in cm^{-1} of compounds [10-16]

Comp. No.	ν(N-H) sulfonamide	ν(C=O) imide	ν(C=N)	ν(C=C) aromatic	ν(C-N) imide	ν(SO_2) asym.	ν(SO_2) sym.	ν(C-O) oxadiazole	Others
10	3407	1710 1660	1612	1585	1355	1375	1157	1190	-
11	3300	1705 1665	1600	1590	1360	1380	1160	1185	**ν(C-Cl) 1090**
Comp. No.	**ν(N-H)**	**ν(C=O)**	**ν(C=N)**	**ν(C=C)**	**ν(C-N)**	**ν(SO_2) asym.**	**ν(SO_2) sym.**	**ν(C-S)**	**Others**
12	3369	1703 1666	1624	1585	1355	1373	1186	651	-
13	3275 3456	1705 1666	1635	1585	1354	1377	1192	655	**ν(C-Cl) 1068**
14	3414	1701 1662	1624	1585 1539	1307	1354	1188	651	-
15	3363 3464	1708 1662	1589	1589	1342	1373	1188	698	-
16	3357	1703 1664	1626	1585	1355	1375	1188	698	**ν(C-Cl) 1092**

Table 3: Antibacterial activity of compounds [10-16]

Comp. No.	Gram-Positive bacteria		Gram-Negative bacteria	
	S. aureus	*S. pyogenes*	*P. aeruginosa*	*E. coli*
10	+++	+++	+	++
11	++	+	+++	+++
12	++++	+++	+++	++
13	+++	++	++	+++
14	++++	+++	+++	++
15	++	+	+	+
16	++	+++	+	+
Ampicilline	+++	+++	+++	+++
DMSO	-	-	-	-

Key of symbol: slightly active = (+) inhibition zone (6-9)mm, moderately active = (++) inhibition zone = (9-12)mm, highly active (+++) inhibition zone (13-17)mm, very high activity = (++++) inhibition zone > 17 mm.

CONCLUSON

A series of new sulfonamido naphthalimides linked to benzothiazole or oxadiazole moieties was synthesized by applying multistep synthesis. The newly synthesized naphthalimides showed good antibacterial activity since their molecules contain three known biologically active components naphthalimide, sulfonamide and benzothiazole or oxadiazole.

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