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Using Nano-Chitosan linked to the drug Naproxen and studying its physical properties and biological effectiveness.

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ABSTRACT:

In this work, the drug naproxen was loaded onto the chitosan nano polymer, and the resulting product was characterized using infrared spectroscopy and proton NMR spectroscopy, and the physical properties of the prepared drug were studied, including its solubility in a number of different solvents, including (deionized water, toluene chloroform, acetone, Ethanol, methyl sulfoxide, hexane,). The absorbance of drug release was measured using UV spectroscopy in two buffer solutions (PH 2.2, 8) at 237°C as a function of time (one hour and a day). The biological effectiveness of the prepared drug was studied by taking two types of bacteria, namely (Staphylococcus aureus) bacteria Gram positive and (Escherichia coli) bacteria Gram negative. The results show that the effect of the prepared chitosan nanodrug gave a much higher result in inhibiting bacteria than using the drug alone (Naproxen). When using two concentrations (1024 and 512) ppm.

Keywords : Nano Polymer , Nano Chitosan , Naproxen , solubility, absorbency, bacteria

Introduction:

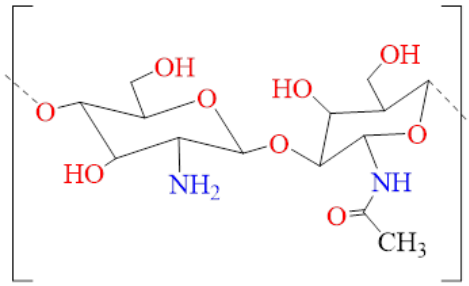
Polymer: Significant advances in polymer chemistry and technology have led to the widespread use of biopolymers, synthetic polymers and their derivatives in the fields of medicine and pharmacy[1]. Scientists have recently paid special attention to medical biopolymers, especially those used in therapeutic systems and drug delivery systems. They have many applications, including masking unwanted tastes, in addition to being the primary tool for regulating the rate of drug release

reducing the number of times you take medication[2] .Large molecules called polymers are composed of smaller molecules (monomers) linked by chemical bonds. Polymerization: The process of attaching a monomer molecule to another molecule is called polymerization[3].

Nano polymer :The Greek word dwarf, meaning little, is where the word nano originates. The study of polymer nanoparticles with particle sizes ranging from 1 to 100 nanometers is the focus of nano polymer science[4]. Their chemical and physical characteristics change when microparticles become nanoparticles, and as a result, they acquire characteristics that make them valuable[5]. Nano polymers are employed in pharmacy and medical in a variety of cutting-edge technological applications to deliver medication to diseased cells without endangering healthy tissues.In addition[6], they aid in the identification of organ cancer and the hastening of wound and skin burn healing[7].

Nano-Chitosan :This polymer is mostly made up of chitin[8], which is taken from the exoskeleton of marine fish like shrimp, oysters, and arthropods[9]. Its molecular weight is (1526.5 g/mol) and its formula is $C_{56}H_{103}N_9O_{39}$[10]. It is an odorless, white to light yellow powder that dissolves in both organic acids and

water. 102.5 C is its melting poin. It is a linear polysaccharide that contains B (1-4)-D-glucosamine. Acetylglucosamine[11]، which is obtained by removing the acetyl group from chitin, According to the equation scales, this process is known as deacetyl[12] ،The following figure shows the structure of nano-chitosan :

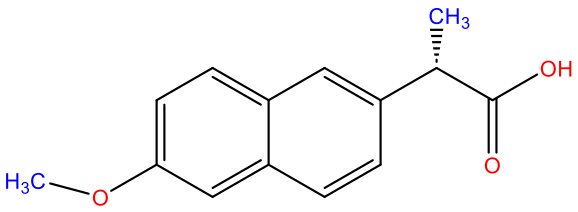


Because of its numerous biological and chemical qualities, including its ease of absorption, antibacterial activity, non-toxicity, and biodegradability[13], chitosan is utilized in medicine for a variety of conditions, including wound healing, infection prevention, and antiacid actions[14]. Due to its great stability, it is also utilized in agriculture and industries. Its non-toxicity and straightforward processing technique have made it useful as a medication delivery vehicle[15].

Drug delivery system : Conjugation of drug to polymer is part of the drug delivery mechanism. This system is characterized by its controlled release[16], which distinguishes it from the traditional method that releases the drug dose into the target area. The treatment continues for the longest period after taking it and travels throughout the body without having any effect until it reaches the target area[17]. The patient benefits from a decreased frequency of drug dosage as well as decreased drug toxicity and fluctuations in plasma level concentration, the escalation of which may lead to additional adverse effects[18].

Naproxen: A pain reliever, which is an opiate derivative similar in structure to codeine, but it is better and stronger in relieving pain[19]. Its molecular chemical

formula is ($C_{14}H_{14}O_3$) and its scientific name is (2-6-methoxynaphthalen-2-yl propanoic acid), It is a white crystalline powder that is practically insoluble in water and dissolves in dehydrating alcohols and chloroform [20]، molecular weight is(230 gm/ mol) melting point (153 C), its biological half-life is (12- 24 hours)[21] , and its chemical structural formula is :



Naproxen is generally used to relieve pain, fever, and inflammation such as arthritis, tendonitis, and menstrual pain (dysmenorrhea) , as well as to treat gout and migraines[22].

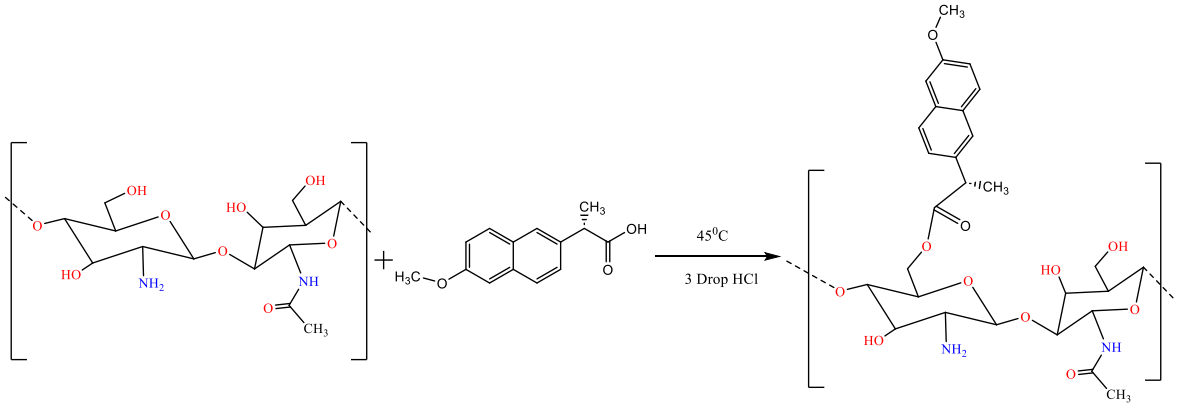
Instrumental & Materials :

Melting points were recorded using a device Electro thermal .VOL 15 Watt .45 W in our university, FT-IR spectra were recorded by using (FT-IR 8300 Shimadzu , Japan) in the range (400-4000) cm- 1 as KBr discs in our university .1H.NMR–Spectra , Bruker – Uitra Shield – 300 MHz Switzerland , with DMSO-d6) carried out in Tehran Universit .UV – Vis spectrometer – (Shimadzu , Japan in our university. An incubator for bacterial growth at the Exir Research Center .Cultivation media sterilization devic Hirayama (HVE- 50) Japan at the Exir Research Center.

Experimental :

Preparation of Nano Chitosan – Drug(N3)

Naproxen drug (2.3025 g , 0.01 moles) were dissolved in 30 ml THF with (3 drops) of concentration HCl and added to nano chitosan (1.5 g, 0.0000034 mole) and reflex for 24 hr. Finally the precipitate was washed by diethyl ether and 2.0 M NaOH and leave to dry for 16 hr.



Physical properties of the prepared nano drug :

The physical properties of the synthesis of chitosan nano medicines were studied, including the solubility of the prepared nanodrug and the drug release characteristics of the nano drug.

1- The characteristics of solubility:

The produced nano-chitosan (N3) was placed in a 0.01 g test tube. After that, attempts were made to dissolve it in a number of solvents, including ethanol, ether, chloroform, DMSO, hexane, and acetone. The solubility of the nano drug in these solvents was then evaluated[23].

2- Pharmacological Release from Nano Chitosan-Drug:

For each type of nano chitosan-drugs that were put in a 50 ml beaker, the amount of drug released from the created nano chitosan-drugs was measured using a UV-Vis Spectrophotometer in two different buffer solutions (2.2, 8) at a constant temperature of 310 K (0.05 gm). For a few days and several hours at a time, absorption (controlled drug release) measurements were conducted.

Preparation of buffer solutions[24]:

Buffer solutions were prepared in the following manner :

1. pH=2.2: This solution was prepared, by mixing 500 ml of 0.2 M of KCl and 8.6 ml of 2.0 M of HCl.

2. pH =8: This solution was prepared, by mixing 500 ml of 0.025 M of

Borax [Na2B4O7.10H2O] and 0.5 ml of 0.1 M of HCl.

Biological effectiveness:

The culture medium was prepared under specific conditions, and the solutions whose biological effectiveness was to be assessed were prepared against two types of bacteria ، namely Gram-positive (Staphylococcus) and Gram-negative (Escherichia coli), at two different concentrations (1024 ppm, 512 ppm)[25].

Results and Discussion:

Organic Investigation N3:

The FT. IR spectrum of compound (N3) Figure (1) shows appearance absorption band appeared at(3446 $cm^{-1)}$ for NH2 and an absorption band at (3377

cm^{-1}) that belongs to the hydrogen bond present in the alcohol (O-H) group, as well as The spectrum showed an absorption band at (3022 cm^{-1}) representing aromatic (C-H), while the band (2895 cm^{-1}) represents aliphatic (C-H) The spectrum showed an absorption band at (1759 cm-1) belonging to the carbonyl ester group. As for the absorption band (1651 cm-1) It belongs to the amide carbonyl group, while the (C-O) band appred at (1261 cm-1)

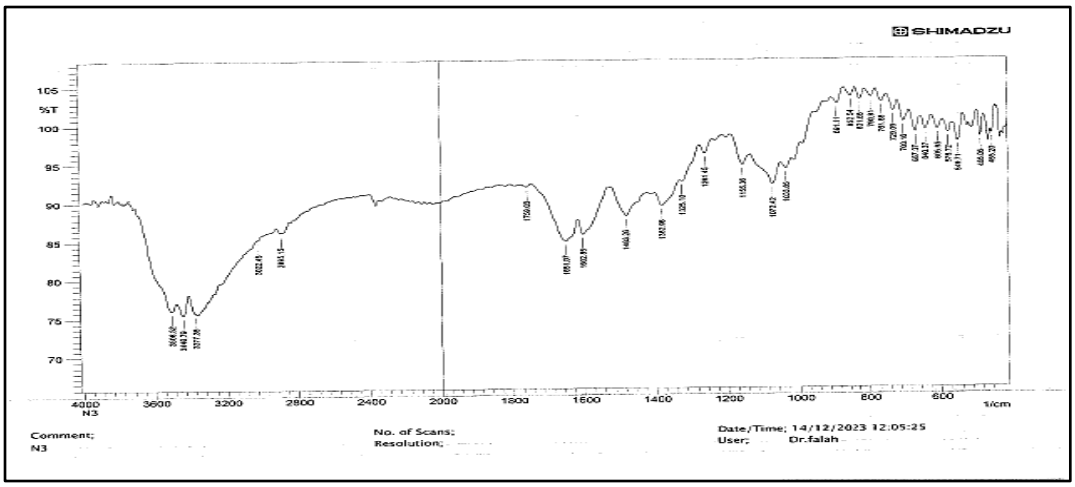


Fig (1) : FT.IR of Compund (N3)

The 1H.NMR where appearance signal of OH group at 8.7 ppm and appear signal of the H (NH amid) at 9.1 ppm and appear signal of the aromatic – H (at 7.2 – 7.8 ppm and appear signal of (CH2OH) at 4.5 ppm and appear signal of (OCH2) at 4.1 ppm and appear signal of(OCH3) at 3.8 ppm and appear signal of DMSO at 2.5 ppm and appear signal of(CH2) at 1.5ppm and appear signal of (CH3) at 1.3 ppm.

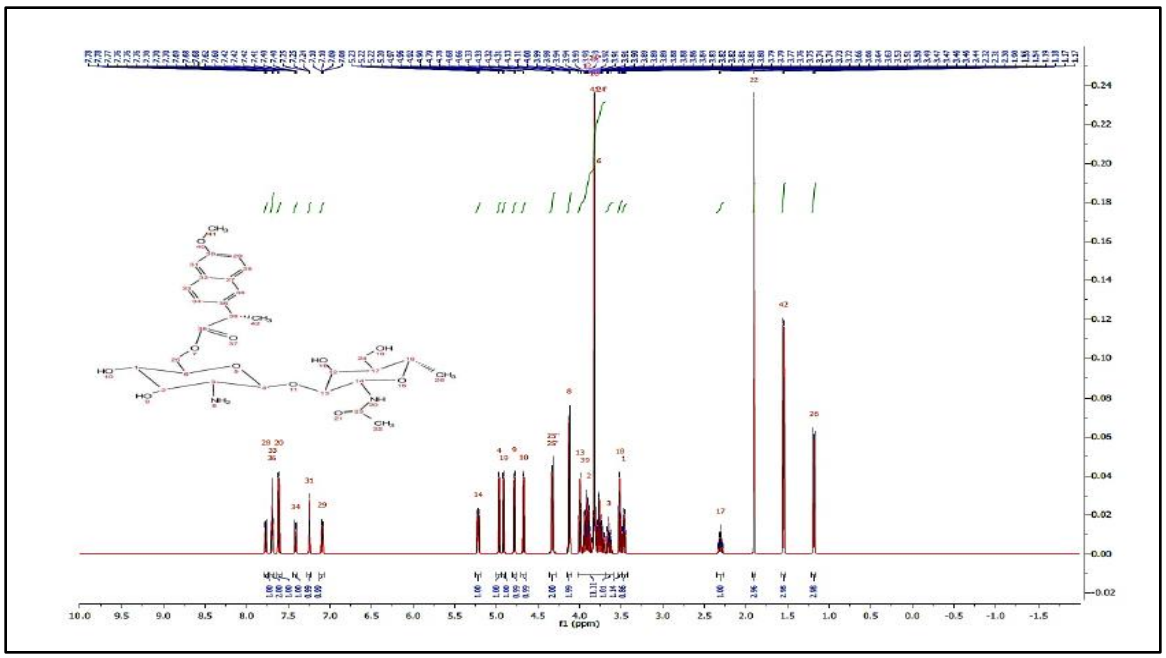


Fig (2) H.NMR of Compound [N3]

Physical properties of the prepared Nano drug:

1- Solubility property: The result of the solubility of the prepared drug in a number of solvents was either completely dissolved(, partially dissolved, or not dissolved at all, according to the following table.

Nano Chitosan drugs	H_2O	DMSO	Acetone	ETOH	Hexane	Chloroform	Toluene
N3	-	+	-	+	-	+	-

The reason for not dissolving in water is that water is ionized and prefers to form hydrogen bonds in order for the dissolution process to occur. Here, the dissolution process does not occur because the size of the drug and polymer molecules is very large, so it does not dissolve in water. As for the dye solvents, hexane, and acetone, they are non-protonated solvents and the size of their molecules is relatively large, so dissolution does not occur As for the solvents, ethanol, chloroform, and

dimethyl sulfoxide are organic solvents that dissolve polymers easily because they are molecularly polar.

Nano drug release:

Using a UV spectroscopy device, the release of the drug prepared from

Chitosan Nano polymer in two buffer solutions (2.2, 8) at a constant temperature of 273C. The release of the drug was measured in hours (5) hours, as well as in days (12) days per pH.

At PH 2.2 :

Time	Nano drug release
Hour	
1	**1.313**
2	**1.355**
3	**1.385**
4	**1.437**
5	**1.437**
Day	
1	**1.502**
2	**1.585**
3	**1.680**
4	**1.774**
5	**1.885**
6	**1.986**
7	**2.120**
8	**2.216**
9	**2.297**
10	**2.369**
11	**2.489**
12	**2.489**

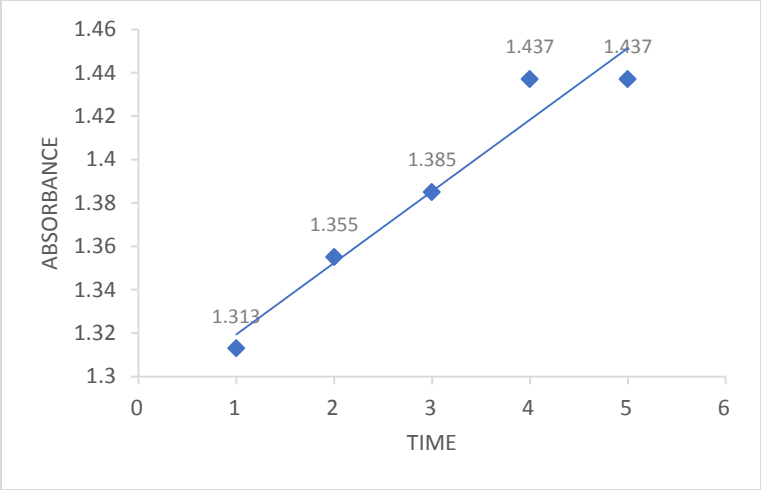


Figure showing hourly drug release curves of chitosan nano drug at pH = 2.2 at 273C

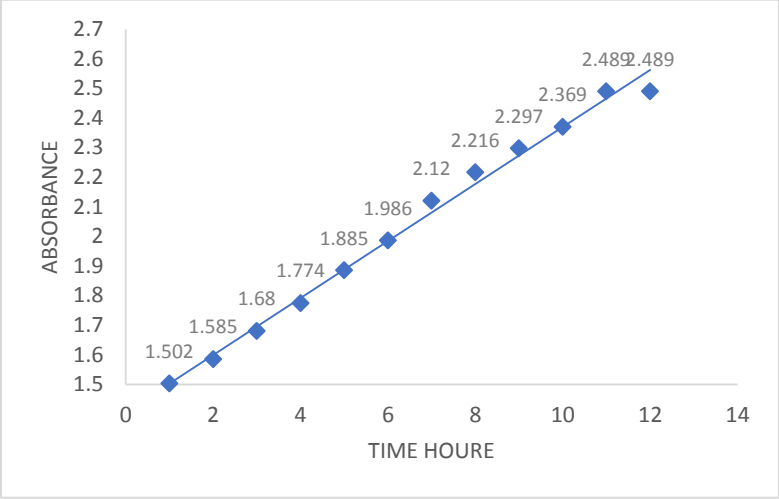


Figure showing drug release curves per day for chitosan nano drug at pH = 2.2 at 273C

From the results of the tables above, it is clear that absorption increases as time increases, whether in hours or days .

At PH 8 :

Time	Nano drug release
Hour	
1	1.555
2	1.597
3	1.637
4	1.700
5	1.700
Day	
1	1.810
2	1.874
3	1.963
4	2.131
5	2.209
6	2.304
7	2.412
8	2.493
9	2.596
10	2.652
11	2.751
12	2.751

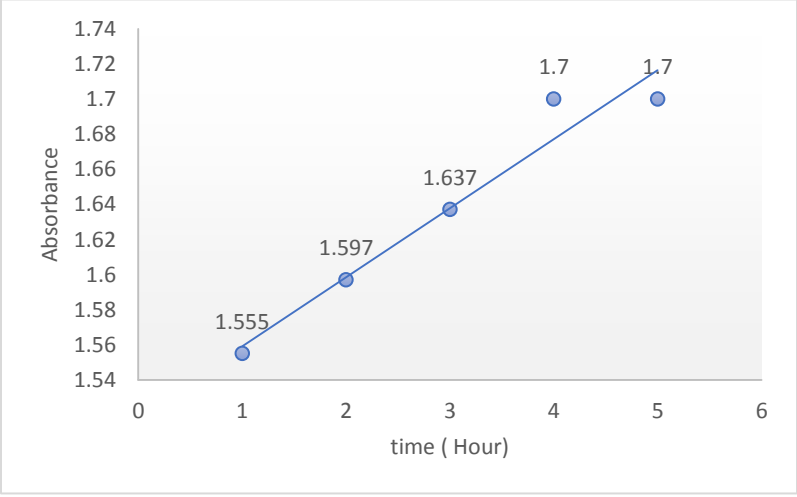


Figure showing hourly drug release curves of chitosan nano drug at pH = 8 at 273C

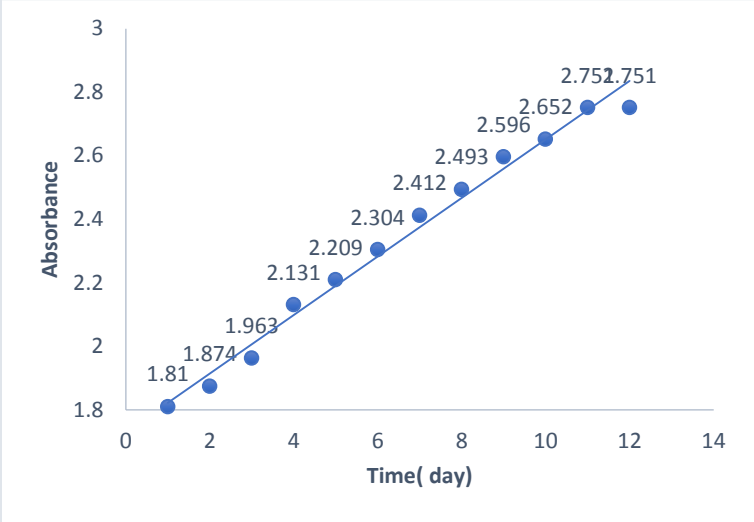


Figure showing drug release curves per day for chitosan nano drug at pH = 8 at 273C

We conclude from what is stated in the tables above that pH has a significant impact on the absorption of the drug, and we note that the absorbance at pH 8 is higher than the absorbance of pH 2.2, and since the measure of pH 8 is the degree of alkalinity of the small intestine, then the process of absorption of this drug will take place in the intestine.

Results of the biological effectiveness of the prepared Nano-Drug Naproxen:

The results showed that the effect of the prepared nano-chitosan drug gave a much higher result than using naproxen alone, as shown below.

Type of bacteria	Naproxen		Drug of nano chitosan (N3)	
	1024ppm	512ppm	1024ppm	512ppm
Staphylococcus	Zero	zero	22	22
Eschershia Coli	Zero	zero	20	20

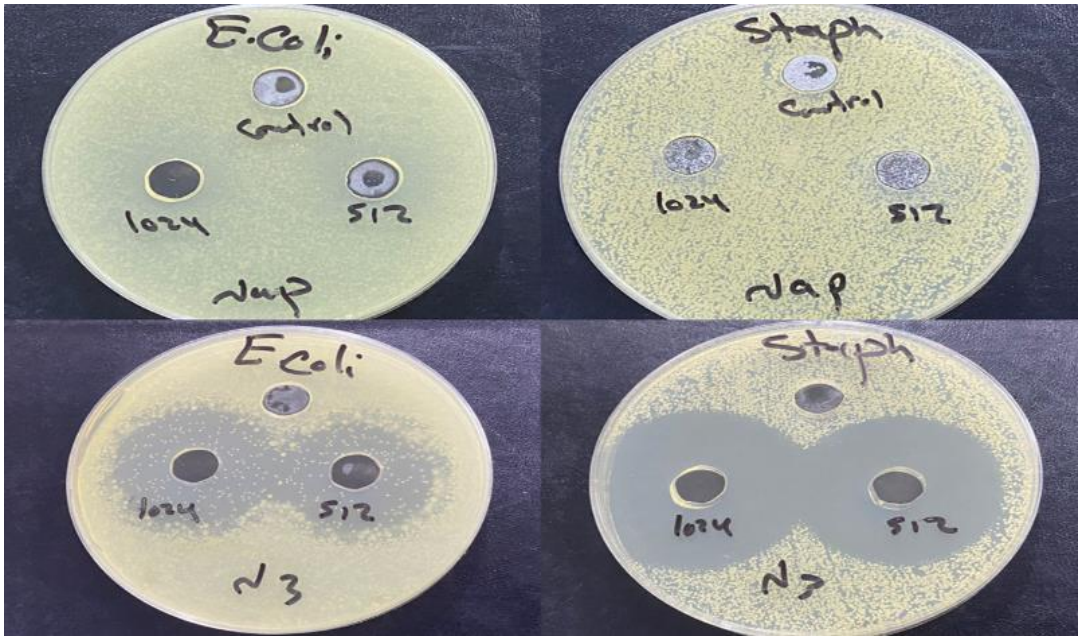


Figure showing the effect of (Naproxen) compared to the prepared drug (N3)

Using naproxen alone did not show any effect on any type of bacteria used, but when the drug was loaded onto the nanopolymer of chitosan, it gave a positive result and was highly effective in inhibiting bacteria**.**

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