Original Article

Extraction, Isolation and Chemical Identification of Piperine Alkaloid from White Pepper Seeds and its Antibacterial Activity

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Abstract

White pepper (Piper nigrum), a member of the Piperaceae family, is a spice plant that is used to improve food digestion and is a crucial component of industrial medicinee. The purpose of this studyy is to demonstrate the antibacteriall propertiess of white pepperr and its active ingredient (piperinee alkaloid), as well as to determine the structure using reagents, TLC technology, and melting point. When compared to raw White pepper and manufactured medications like cefuroximee, piperine alkaloids exhibited a good biological activity against E. coli, Staphylococcus aureuss, Pseudomonass, Streptococcus, Aeromonas, Klebsiellaes, and Acinetobacter.

Keywords: White pepper, Piperine, AntibacteriallActivity, Isolation, and Identifications.

1. INTRODUCTION

The family Piperaceae's main genus, Piper, is home to moree than 3,000 species that have been foundd throughout tropical and subtropical areas of the world. China is the home to about 60 species, 12 of whichh are found on the island of Hainan. The most significant commercial plant in China was pepperr (Piper nigrum L.), which was brought to Hai from Indonesian in 1947. Although there are more than one hundred different varietiess of peppers known to exist, only a small numberrof them are widely used for human foodd. Pepper cultivars were developed from wild species [1].White pepper has therapeuticcc properties because it containss piperine, an active substance with the chemicall formula C17H19NO3 .

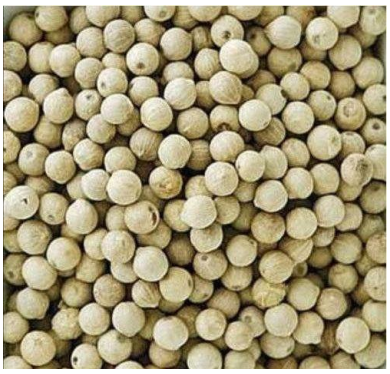


Figure No 1: white peppers

white pepper containss starch, 6% pungentt resin, 2.5% essentiall oil, and 4.6–9% piperine. White pepper oleoresin extracts have been shown to have anti-inflammatory, depressivee, analgesic, anti-rheumaticc, and adiabatic capabilities qualitiess.the piperine is the actives constituent of white pepper.

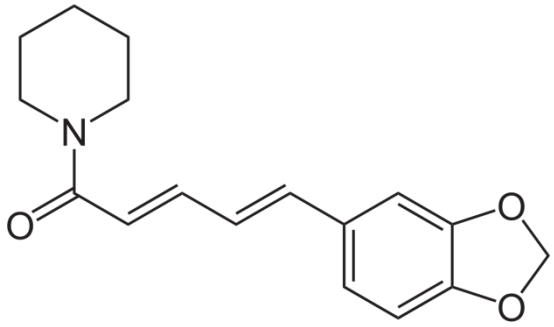


Figure No 2 : It shows the structure of piperine

The purpose of this study is to identifyy a combination of operating circumstancess for an ideal extraction yield of spice oleoresinn from white pepper employing taguchi orthogonal design becausee of its capacity to demobilize. White pepper as shown in figure No1[2] .When used orallyy, white pepper has several undesirablee side effects, such as stomach distress and gastroo intestinal issues. This can be due to the medicinee being available in its entiretyy at one moment. A sustained releasee method for the medicine to remain in the therapeuticc window over the course of the therapyy might be a good concept in this regard. Recent studies havee focused a lot on using natural polymers as drugg carriers to maintain actionn. Here, sodium alginate is employedd as the polymer and the ionotropic gelation processs is used to create micro beads. By creating an eggf box junction to link the divalent metall ions of the alginate polymer chain 6, the gelationn is brought about [3].

2. MATERIALI AND METHODS:

2.1 Materialss:

White pepperr seeds were bought in Nasiriya's local farmers market, pounded into a powdery and then stored in a dry jar All substances were sourced from a college lab The work was done in the lab for organicc chemistry.

2.2 Extract and isolating techniques for piperine alkaloids :

In this work, piperine alkaloids from white pepper were extracted and isolated. In order to remove non-polar elements from the white pepper plant, suchh as aromaticty oill, waxess, and resinss, the maceration extraction was made by combining 120 g of white pepper powder with 250 ml of petroleum ether. 120 gm of white pepper powder and 200 mllileter of 90 % ethanol were combined to create the ethanolic extract, which was then extracted for two hours using a reflux extractor condenser. The extracted solution was then collected and chilled. The extract solution was then concentrated to 30 ml and heated to 600C. then 3 mllileter of 15 % Alcohol KOH was added to the concentratedd white pepper solution to precipitate the acidicc resins, followed by 3 hours of filtering before the alcohol extract was allowed overnight to produce yellowish brown needles with a melting point of 1240C. 0.8 g of piperine alkaloids were produced [4-8].

Chemical Reaction Scheme of extraction and isolation of Piperine

First step : 50 gm of Blackx Pepper

150 ml of petroleum ether

Maceration extraction (defatted process)

Second Step : 50 gm of Black Pepper

100 ml of 95 % C_2H_5OH

3 hrs Reflux extractor

Hot filtration

Heating Concentrated

Third Step : 100ml of extract solution on water bath at 600C

3 ml of 10 Alcoholicc KOH

Fourth Step : Filtrated to remove the Co-precipitate of resins acids

left over night

Scheme No 1 extractionn of piperinee ((Piperinee Alkaloid)) fromm black pepperr

3.Results and Discussion:

3.1Chemical Identificationn of Pure Piperinee Alkaliodss

3.1.1Quantitative Analysiss: This was carried out by weighing the crystalss of the alkaloid piperinee .

Results: needles in yellow as shown in the figure No 3 0.9 grams of piperinee alkaloids were produced .

Figure No 3: Yellowishh brownnneedles of piperinee

3.1.2 Analyses of the qualitative kind (chemical identification)

Following their chemical identification by the subsequent tests and TLC/melting point, piperine alkaloids were extracted of the acid resins and others polarr elements present in the white pepper using the method depicted in above Schemee No. 1[8-15].

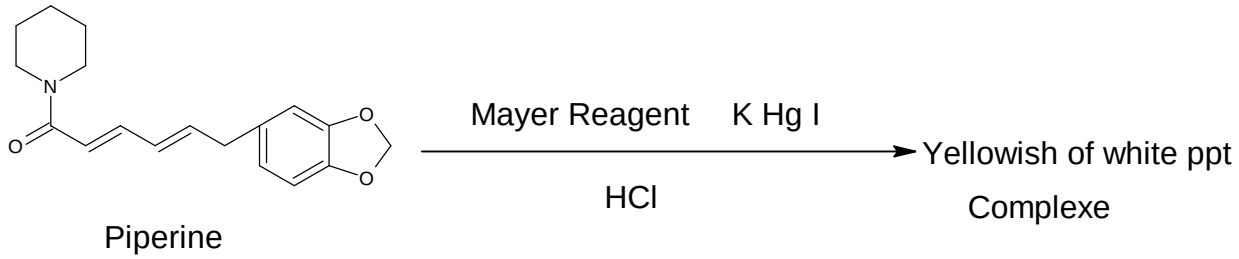
1. Test of Mayer 's:

The majoriteis of alkaloid are precipitated of neutral or slight acid solution by Mayer reagentt (potassiomercuric iodide(I2) solution to gives ceramic to off white a colored precipitatee. (Mayer reagentt is freshlyy synthesized by dissolvings a mixtures of (HgCl2)mercuric chlorided (1.38 gram) and of potassium iodided(KI) (6.00 gram) in distillation water (D.W) (150 milliter).

Procedure :

Add two drops of HCl to a Petri plate after dissolving a fewest crystals of piperines alkaloidd in a little amount of ethanol . Next, put 2 drops of the solution in.

Result : a precipitate that was yellowishh off white developed.



Scheme No 2 : (testr of Mayerr)

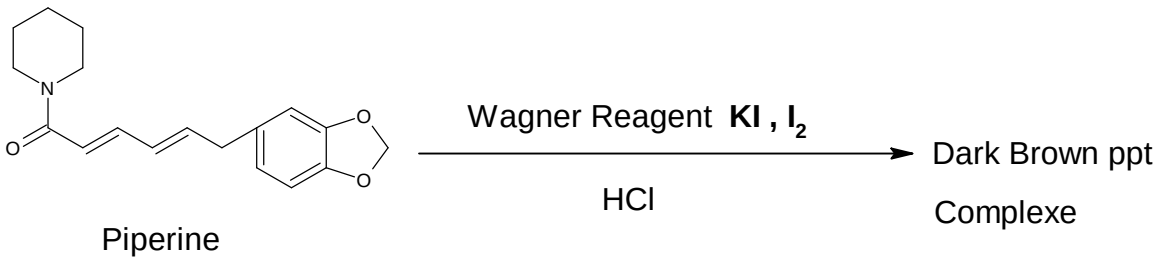
2. Test of Wagner:

Aim of test: to generalize the alkaloid and compare it to other alkaloids.

(Dissolving 3 gm of Iodinee and 8 gm of potassium iodide KI in 100 ml of D.W)

Procedure: Add two drops of HCl to a Petri plate after dissolving a fewest crystals of piperines alkaloids in a little amount of ethanolic. Add 3 drops of reagentt of Wagner's after that.

Result: a dark brown precipitation appeared.



Scheme No 3 (test of Wagner)

3.Chromatography (TLC) :

Following its separation, piperine was identified using the chromatographic techniques TLC and Silica gel G. As stationary, ethanol:hexan 7:3 is the mobile phase, while dragendorff's reagent is the spray reagent. The literature's conventional Rf value for piperinee was 0.26. The refined Rf- valuee. TLC piperine was determined to contain 0.23[16-25] .

Result : An orange spots will emerge.

4. Meltingg pointt :

point of melting of Piperines Alkaloids 1230C, which from the literature seemed to be near to Piperine Melting Point 1250C.

5. Making a stock solutionn:

The stocks solute of White pepperr and the actives ingredientt piperine was used for tests no. (1-4), wheree 0.2 gramm of white peppers and the actives ingredient piperine were dissolved in 30 millileter of ethanoll to produce a concentrations of 6 mg/mll, whichh were the concentration testedd, and no. (4-7) where 0.4 gm of white pepperr and the active ingredient piperines were dissolvedd in 10 mllileter of ethanoll to produce a concentrationn of 41 mg/ml, which was the concentrationn (No 1). Filtering the items via a Millipore 0.46 mm and 0.23 mm cleaned them[[26-30].

6. Biochemical action:

The activty ingredient (piperines) were chemically removed & separated from white pepper, and the anti-bacterial activity of white pepper were both assessed in vitro to use the agar cups technique. Staphylococcuss, E. colii, aureuss, pseudomonas, streptococcuss, aeromonass, klebsiellaa, and acinetobacterr were evaluated for activity against all of the compounds. According to the information in table (No1), black pepper's active ingredient (piperine) was more effective than a piperine [31-36] against the pathogens that were chosen, with the exception of test (No7), which revealed that black pepper was more effective than a piperine against Acinetobacter. Table No. 1 provides a summary of the findings.

Table 1 : shows the diameter zone of growth of bacteria inhibition (mm) by plant extracts .

Types of Isolates	Antimicrobial Agent			
	CXM30	Plant extracts		
		General White Pepper	Piperine	Test No
Staphylococcuss aureus	19 mm	5mm	28mm	I
E. colii	R	1 mm	7 mm	II
Pseudomonass	R	1 mm	8 mm	III
Streptococcuss		27 mm	31mm	IV
Aeromonass		19 mm	27mm	V
klebsiellaa		19 mm	41 mm	VI
Acinetobacterr		39 mm	12 mm	VII

Acinetobacterr 39 mm 12 mm VII

CXM= CEFUROXIMEE

4.CONCLUSION:

As discussed previouslys, Isolate Piperine alkaloidss which froing white pepper seed were charecterzied by chemical test of

Waagner , Maayer , point of melt 1260C and Thin layaer chromatography(TLC) . resultss of pharmaceutical acvtivites study suggest that the ethanolic extract of Piperines alkaloidal found very much effectivly against Escherichia coli E. coli , Staphylococcuss ,aureeus , Pseudomonaas, Streptococcuus , Aeromonaas , klebsiella ,Acinetobacteer, Comparable to the crudes white peppers as seen in inhibition zonee excepted Acinetobacteer which wase shown very good zone inhibition with crudes white pepper.The most importance in our studies the Piperines alkaloids which are extracted and isolated from white pepper are show more effective and good abiological activity than the prepared drugs CXMe = cefuroxime .Piperinee to examine the different pharmacological activities of like tetrazine, oxadiazole, piperine, harmalole, and other heterocycles, different synthetic and natural heterocycle substituents were used which are prepared by author and fond have good antibacterial activity and good pharmaceutical activy [14–36].

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