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# Medicinal Activity of Alkaloidic Compound 1H-pyrido{2,3-b} indole Isolated from Solanum melongena Cortex Against Some Pathogenic Bacteria

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**Abstract**
Current study was carried out for extraction ,isolation ,separation and identification of the alkaloidic compound which is represented by (1H-pyrido**{2,3-b}** indole from Iraqi *Solanum melongena* cortex by using cold ethanolic extract ,qualitative analysis ,quantitative isolation and gas-chromatography –mass spectrum(GC-MS) technique. This active compound was applied for estimation and investigation of its medicinal activity by using various concentration agains two pathogenic bacteria which were represented by *Escherichia coli* and *Staphylococcus aureus* .the concentration (10, 15, 25. 50, 100 and 200 mg/ml),recorded inhibition zones diameters equal to b(20,21,22,21,23 and 41)against growth of *E.coli* bacteria strin wheras the same concentrations showed inhibition zone diameters equal to (1.0,1.0,25,30 and 43 mm) against growth of *S.aureus* bacteria strain the minimal inhibitory concentration was 0.1 gm/ml for both bacteria strains .Therefore (1H-pyrido(2,3,-b ) indole compound can be used for treatment of different diseases caused by these pathogenic organisms but this research demands further clinical and pharmaceutical studies .
**Keywords**: *Solanum melongena* cortex, 1H-pyrido**{2,3-b}** indole ,pathogenic bacteria ,GC-Mass technique .medicinal activity .

## 1. Introduction

Medicinal plants have a great importance to the health of individuals and communities because they have biochemical ability to treat several various diseases such as diabetes mellitus ,hyperthyroidism and cancer. The medicinal value of these plants comes from presence of different active phytochemicals abundant in various parts of medicinal plants (Nduche *et al.,*2015;Abed Al-Majeed *et al*.,2016) .Recently several phytochemical studies were achieved that concerning natural chemical compounds existing in medicinal plants which are represented by phenols ,alkaloids ,glycosides ,terpenes ,steroids and saponins having medicinal effects as successful remedies to treat various diseases and also use theses phytochemicals instead of antibiotics that have many side effects(Osadebe *et al.*,2014; Pramila et *al.,*2012). The search on active phytochemical metabolites is now happening in order to investigation their biochemical potential lead to provide new natural drugs which have no side effect ,therefore herbal plants were used usually in traditional medicine in many countries since they showed positive results in treating different diseases . Several active chemicals families were isolated from different medicinal plants such alkaloids from *C.rutidosperma* , tannins from *S.dulcis* ,flavonoids from *T.procumbens* ,saponin from *S.acuta* ,steroids from *S.anthelmia* and terpenoids from *E.coccinea* plants (Edeoga *et al*.,2005; Ok wu and Josiah .,2006)) .*Solanum melongena* is an economic flowering plant which. belong to family *Solanaceae* and this family has seventy five genera and over 2000 species .Members are mostly herbaceous plants and its fruits are berry and the seeds large endosperm and they are grown mainly as food and drug .Many studies reported that this plant have various phytochemical compounds such as flavonoid s, terpenoids , tannins ,glycosides ,steroids and alkaloids (Saleh , 2015; Amer and Abdelmohsen .,2014).

Alkaloids are nitrogenous heterocyclic compounds have basic properties because presence of nitrogen atoms and some of them have neutral and weakly acids features . In addition of carbon, hydrogen and, this active class may also contain oxygen . sulpher and rarely other elements such as chlorine, bromine and phosphorus .Alkaloids as phytochemical active compounds are abundant in different medicinal plants such as gibberlin in *Gibberella fujikuroi*, atropine in *Atropa belladonna* and codeine in *Papaver* some nifoerum (Kabera *et al.*,2014;Sun,2010;McBrien *et al.*,2013;Simera *et al*.,2010).The alkaloids compounds are biochemically synthesized by using a great various number represented by organisms include bacteria ,fungi. animals and mostly by medicinal plants as secondary metabolites .Different classes of alkaloids depending on similarity of the carbon skeleton, and all of them are biochemically produced from amino acids substrates such as tyrosine and phenyl alanine (Mohammed and Al-Maliki,2014; verpoorte,1998). The current study was aimed to investigate the medicinal activity of an alkaloidic compound isolated and identified from *Solanum melongera* cortex against growth of some pathogenic bacteria depending on biochemical effect in increase of inhibition zone diameters then killing these micro organisms.

## 2.Materials and methods

### 2.1. Study plant

*Solanum melongena* (egg plant ) fruit were purchared from Garmate Ali local market in Hartha district at Basrah governorate in Iraq. The cortex of plant was isolated from fruits , cleaned by distilled water, dried ,ground by electrical mill and kept in dark glass containers in laboratory until the use . this plant was taxonomied in biology department in college of education for pure sciences at university of Basrah -Iraq by a specialist botanist .

### 2.2.Preparation of cold alcoholic extract of *Solanum melongena* cortex .

Twenty five grams of ground cortex of the plant were mixed with 500 ml of ethanol solvent in conical flask and the mixture was shaken well , then it was stirred on magnetic stirrer for 10 hours . After that the mixture was filtered by using Buchner funnel the precipitate was removed and the filtrate was collected and dried as crude (Harborne,1984).

### 2.3. Preliminary Qualitative and Analysis of alkaloids in ethanolic Extract.

Alkaloidic compounds were tested in the cortex ethanolic extract by using Dragendroff s reagent where 0.1 gm of crude was dissolved in 1 ml of ethanol then 2.0 ml of this reagent was added to the soluble crude and the mixture was shaken well .After that orange precipitate was formed (Harborne and Bxter,1993).

### 2.4. Identification of cold ethanolic extract by GC-MASS technique

The cold ethanolic extract of *Solanum melongena* cortex was separated and identified by using gas chromatography mass spectrum (GC-MS) technique in collage of agriculture at university of Basra in Iraq by using shimadzu-GC-MS –Qp 2101 ultra instrument .

### 2.5. Isolation of alkaloids from *Solanum melongena* cortex

Twenty five grams of ground cortex of the *Solanum melongena* plant were mixed with 500 ml of ethanolic acetic acid (10%v/v) and the mixture was stirred on magnetic stirrer for twelve hours then it was filtrate by Buchner funnel .the precipitate was removed and the filtrate was concentrated to quarter of its volume then 5 ml of sulphuric acid added to the filtrate and the content were mixed well . After that pH was adjusted to 9 by adding 4 ml of ammonium hydroxide and the basified solution was put in separation funnel then 20 ml of chloroform for 3 times was added to alkaloids extract . The extract was mixed well and two layers were formed as organic and aqueous layers .then alkaloids were separated from organic layer as crude (Goadwin and Mereer,1993 ;Cowan ,1997).

### 2.6. Pathogenic Bacteria

The pathogenic standard bacteria strains were isolated and identified which are represented by *Escherichia coli (*negative towards Gram s stain) and Staphylococcus aurous (positive toward Gram s stain).

#### 2.6.1.Culture medium

Muller Hinton agar medium was prepared according to full information determining by manufacturing company and this medium was gotten form marine chemistry department in marine sciences centre at university of Basra-Iraq.

### 2.7. Estimation of medicinal activity of alkaloidic compound

Agar –diffusion method was used by adding 0.2 ml of Muller –Hinton agar culture medium for each glass plate then 0.1 ml of bacterial suspension hasing 0.1 optical density with wavelength equal to 450nm was added into medium by using spectrometer . The concentrations (10, 15 ,25 , 50, 100 and 200 gm /ml ) for alkaloidic compound were prepared and treated against growth of pathogenic bacteria represented by *E.coli* and *Staphylococcus aureus* to determine the inhibition zone diameters .After that these concentrations and bacteria were put together in the incubator at 37 C for 24 hours , finally the antibacterial activity for each concentration was measured (Chauhan *et al,*2010).

## 3. Results and Discussion

The weights cold ethanolic extract and the alkaloidic compound isolated from *Solanum melongena* cortex were found equal to 3.25 and 2.19 gm respectively , therefore the extraction percentage of these extracts are in dictated in table (1) .

**Table (1) Extraction percentage of cold ethanolic and alkaloidic extracts**

| No. | Extract type | Ground plant weight (gm ) | Extract weight (gm) | Extraction percentage |
|---|---|---|---|---|
| 1 | Cold ethanolic | 25 | 3.25 | 13% |
| 2 | Alkaloidic | 25 | 2.19 | 8.76% |

From table (1),it was noticed that the cold ethanolic extract has more extraction percentage (13%) than

alkaloidic extract (8.76%) ,so this means presence of many active chemical compounds in *Solanum melongena* cortex in addition abundance of alkaoidic compound . also the extraction percentage of the active alkaloid was very good compared with cold ethanolic extract percentage , therefore this fact ensure presence of alkaloid quantitatively and qualitively in *Solanum melogena* cortex. The existence of so the presence of these active chemical compounds, alkaloidic compound as active phytochemical in medicinal plants including eggplant indicates the high medicinal activity of these plants .

### 3.1. Qualitative analysis results of ethanolic extract .

Table (2) represents the results of qualitative test of the alkaloidic compounds in the cold ethanolic extract of *Solanum melongena* cortex . By using Dragendroff s reagent towards this extract , the orange-red precipitate was formed cleary in a high quantity , therefore this positive result indicates presence of alkaloids in this medicinal plant (Kim *et al,*2010;Zha *et al*,2010; Shenta and Al-maliki ,2013).

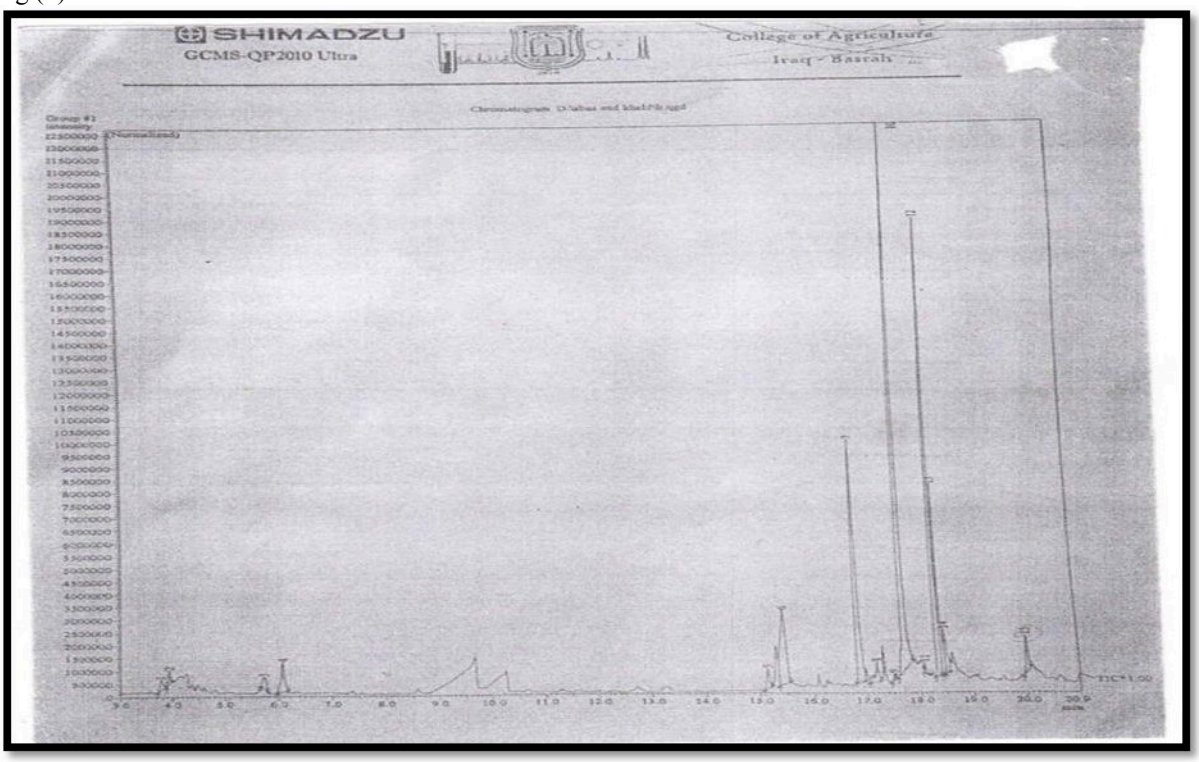
**Table (2)Qualitative test of cold ethanolic extract prepared from *Solanum melongena* cortex**

| Extract type | Reagent | Test | Notice | Conclusion |
|---|---|---|---|---|
| Cold ethanoic | Dragendroff | +++ | Formation of red-orange precipitate | Presence of alkaloids |

The presence of alkaloids as active natural compounds in medicinal plants led to isolate them as pure chemical compound ,separation of them and identification of all these phytochemicals to know their chemical structures then use of them as herbal drugs in various medicinal fields .Several studies illustrated abundance of alkaloids as active compound in different medicinal plants such *Cinochona succirubra ,Xanthorhiza Simplicissima* and *Tinospora Cordifolia* (Gu *et al*.,2011; Mwita *et al.*,2012).The biochemical importance of presence of alkaloids in various medicinal plants ,results from their effects as therapy for different diseases and also they have antimicrobial activity (Surya and John,2001).

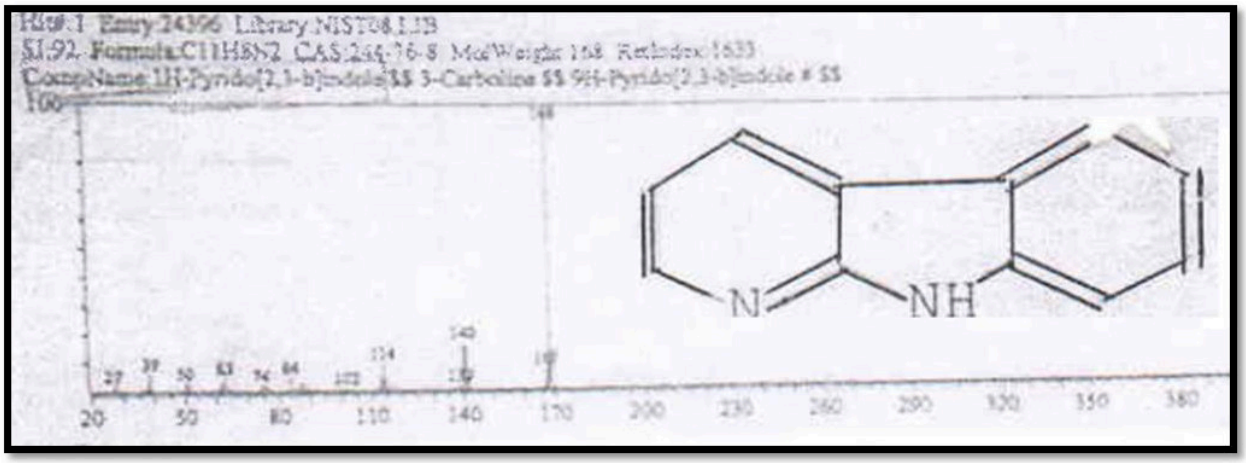
### 3.2. Gas chromatography –mass spectrum technique results

Several different chemical compounds were separated and identified by using GC-MS spectrum technique depending on their various peaks and related mass spectra . From these phytochimcal , one alkaloidic compound was separated by gas chromatography with a peak no.9 hasing a retention time equal to 17.518 min as in fig.(1)




**Fig(1) Chromatogram of chemical compounds separated by GC-MS technique**

Then active compound was identified by mass spectroscopy and from data information of gas chromatography and mass spectrum , it was found that the alkaloid separated and identified is (1-H-pyrido {2,3-b} indole as in the fig(2) .




**Fig(2) Mass spectrum of (1-H-Pyrido{2,3-b} indole alkaloidic compound identified by GC-M S technique**

Therefore GC-MS technique was carried out successfully in separation and identification of the alkaloidic compound including of the compound represented by (1-H-Pyrido {2,3-b}) indole as a natural phytochemical then this chromatographic and spectral tool provides a fantastic and characteristic indications to get the chemical identity and fine structure of all active chemical compounds including alkaloids (Khan,2010).

Therefore(1-H-Pyrido{2,3-b}) indole was isolated from solaunum melongena cortex by using ethanolic acetic acid (10% v/v) solvent which has ability to isolate alkaloids from other active chemical compound . Ammonium hydroxide was added to mixture containing alkaoildic compound to adjust the pH to value equal to 9 . Chloroform was agood extractor for the active alkaloidic compound . Many pre-studies ensure the high performance of chloroform as an extractor for alkaloids existing in the medicinal plant such as *Albizia lebbeck* (Uma *et al*.,2009 ; Al-maliki,2011).

The biochemical benefit of abundance of alkaloids in medicinal plants is to islolate the toxic materials from plant , storage of some essential elements such as nitrogen ,regulators of growth and protecting the plant from attack of fungi and insects (Borner and Varner,1969), Figure(3) shows the fine chemical structure of active compound (1-H-Pyrido**{2,3-b}**) indole.

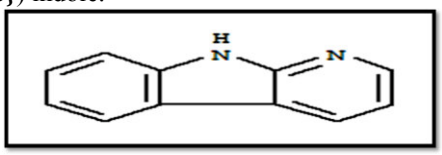



**Fig.(3) structure of (1-H-Pyrido{2,3-b}) indole isolated from *Slaunum melongena* cortex**

### 3.3. Medicinal activity of (1H-pyrido{2,3-b})indole compound .

The medicinal activity for alkaloidic compound which is represented by 1H-pyrido **{2,3-b}** indole isolated from *solanum melongen*a cortex was studied by using several concentration against tow pathogenic bacteria. The concentrations (0.01,0.015, 0.025 , 0.05, 0.10 and 0.20 gm/ml) recorded the inhibition zone diameters equal to ( 20 , 21 . 22 , 21 , 23 and 41) against growth of *Escherichia coli* bacteria , whereas the concentrations ((0.01,0.015, 0.025 , 0.05, 0.10 and 0.20 gm/ml) showed the inhibition zone diameters aqual to (1.0 , 1.0 , 1. 0 , 25 ,30 and 43 mm ) against growth of *Staphylococcus aureus* bacteria as in tables (4 and 5), the minimal inhibitory concentrations were found to be 0.01 and 0.025 gm/ml for *E.coli* and S.aureus bacteria respectively . it was noticed that the increase of concentration led to increase of inhibition zone diameter then increase of medicinal activity . Also the increase of inhibition process is explained by ability of active chemical compound 1H-pyrido **{2,3-b}** indole to kill more pathogenic bacteria .

**Table (3)Medicinal activity of 1H -pyrido {2,3-b} indole compound isolated from *Solanum melongena* cortex against *E.coli* bacteria**

| Alkaloidic compound | Pathogenic bacteria type | Alkaloidic compound conc. (mg/ml) | Inhibition zone diameter(mm) |
|---|---|---|---|
| 1H-pyrido {2,3-b} indole | *Eschrechia coli* | 10 | 20 |
| | | 15 | 21 |
| | | 25 | 22 |
| | | 50 | 21 |
| | | 100 | 23 |
| | | 200 | 41 |

**Table (4)Medicinal activity of 1H -pyrido {2,3-b} indole compound isolated from *Solanum melongena* cortex against S.*aureus bacteria***

| Alkaloidic compound | Pathogenic bacteria type | Alkaloidic compound conc. (mg/ml) | Inhibition zone diameter(mm) |
|---|---|---|---|
| 1H-pyrido {2,3-b} indole | *Staphyllocaccus eureus* | 10 | 1.0 |
| | | 15 | 1.0 |
| | | 25 | 1.0 |
| | | 50 | 25 |
| | | 100 | 30 |
| | | 200 | 43 |

The biochemical mechanism of the high activity of this alkaloid belongs to chemical bonding between this alkaloidic phytochemical with nucleic acids (DNA and RNA). Then inhibition of metabolism of these acids in the cell of these pathogenic microorganism (Jayasurriya *et al*.,1991,Pattnaik and Sharma ,2004). Alkaloids also denaturant the living cell proteins and they an interaction with enzymes of protein biosynthesis containing thiol group (-SH),Active alkaloidic compounds are capable of linking with enzymes in protein metabolism such as DNA-polymerase and RNA-polymerase (Shah *et a*l.,2006 ) . Also imine group (-N=C ) as functional group has a higher activity towards both of pathogenic bacteria , therefore the medicinal activity increase with increase of the number of these active groups (Hollman and Katan ,2001).

## 4. Conclusions

The active alkaloidic compound represented by 1H-pyrido**{2,3-b}** indole which was isolated and identified from Iraqi *Solanum melongena* cortex showed an excellent medicinal activity against growth of *E.coli* and *s.aunreus* pathogenic bacteria where it recorded great inhibition zone diameters towards these bacteria especially at the high concentrations then it has characteristic ability to kill most of these bacteria. Therefore this alkaloidic compound can be used as therapy for various disease caused by these bacteria but this work demands further clinical and pharmaceuitical studies.

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