GC-MS ANALYSIS REPORT ABOUT PUMPKIN FRUIT *(Cucurbita maxima, Cucurbitaceae)*

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ABSTRACT

The pumpkin is a gourd of the genus *Cucurbita* and the family Cucurbitaceae, it is refers to cultivars of any one of the species *Cucurbita pepo* or *Cucurbita maxima*. The study revealed that pumpkin fruit studied was a potential food and has a multiple components of medicine benefits to the weak patients. Twenty two bioactive compounds and their important biological activities were degraded from ethanolic extract of pumpkin fruits by using GC-MS analytical technique. However, further studies should be done for isolation of individual phytochemical content and their biological activity will definitely give fruitful results.

**Keywords:** pumpkin fruit, GC-MS, ethanolic extract.

INTRODUCTION

The pumpkin is a gourd of the genus *Cucurbita* and the family Cucurbitaceae, it is refers to cultivars of any one of the species *Cucurbita pepo* or *Cucurbita maxima* (Cred, 2008).

They typically have a thick orange or yellow shell, creased from the stem to bottom, they have a moderately hard rind with thick, edible flesh below and a central cavity containing numerous seeds (Credo, 2008). Although pumpkin fruits are usually orange or yellow, some fruits are dark green, pale green, orange-yellow, white and grey and can be stored without damage for a long time and that due to the thickness of it is wall (Dhiman et al, 2012).
Pumpkin in Quran & Sunnah

Pumpkin mentioned in Quran Allah says :(( and we caused by a tree of gourd to grow over him)) (Al-saaffat, 146), as the wisdom of the god in the pumpkin to germinate pumpkin plant on the prophet Yunus for food and healing.

The Mufassirin (commentators) said: The gourd (al-Yaqtin) is a kind of pumpkin. Some of them described the benefits of the pumpkin, such as: it grows quickly, it provides shade, it has large, smooth leaves, it keeps flies away and its fruit provides good nourishment: it can be eaten raw or cooked. It is known that the Messenger of Allah (peace and blessings be upon him) liked this kind of pumpkin and used to look for it on the plate of food (Tafsir Ibn Kathir). Ibn Al-Jawziyyah for pumpkin fruit : (it cuts thirst, relief headache, laxative, and is one of the nicer foods, the seeds of pumpkin expel tape worm from intestine, and good for patients with kidney problems, bowel, arthritis and rheumatism). The prophet Mohamed (ppbuh) said : (it is strengthens the mind and the brain) (Albukhary, 5379).
Popularity of pumpkin in various systems of traditional medicine for several uses as:

- Sedative, emollient and refrigerant; used as poultice, applied to burns, inflammations, boils, and burns (Susan D Van-1998).
- The pumpkin seeds and fruit increase power of intelligence and mental vitality (Carbin BE-1990).
  - Gastro duodenal protective and anti-ulcerogenic (Sarkar & Buha, 2008).
- The fruits of pumpkin are mildly natural diuretic and have been used in Central American medicine as a treatment for nephritis and other urinary system problems (Caili et al., 2006).
- Lower cholesterol blood level (Phillips KM, 2005).
- Pumpkin fruits pulp and leaves is used as external emollient and anti inflammatory to soften the skin dryness or roughness, pimples, spots, freckles.
- Anticancer agent and Anti-oxidant activities (Liu et al., 2008).
- Pumpkin fruit has a laxative action, then being useful in case of gastrointestinal disorders such as dyspepsia and constipation (Tylor-1993).
  - Anti-diabetic agent (Quanhong et al., 2005).
- Antihypertensive agent (decrease elevated blood pressure) (Zhang X-1994).
- Antimicrobial agent and for intestinal inflammation (Cowan, 1999).

Objectives

1- To investigate its nutritive value.
2- To know its phytochemical components.
3- To illustrate the major components.

Materials and Methodology:

Materials:
plant:

A pumpkin Fruit was purchased from the local market of Khartoum city in February 2012; authenticated, dried at shade and reduced to fine powder using pestle and mortar in the laboratory as described by (Bean, A.R, 2006). The powder was stored dry, and used as the stock sample for further analyses.

Instruments and chemicals:
A wide range of instruments and chemical were used during the course of this study.

**Method:**

In this study all experiments were divided into four main parts:

1. Extraction of all active constituents in pumpkin fruit using ethanol as solvent.
2. Phytochemical screening test and antimicrobial activity tests.
3. GC-Ms analysis for pumpkin fruit extract.
4. The structure activity relationship and correlation between the detected active constituents and the traditionally reported uses of the pumpkin fruit.

**Generally, GC analysis conditions:**

We were likely need to optimize GC conditions depending on their analytical needs, equipment, and sample type. In general, a typical gas chromatography method will include:

- Column: HP-5 MS 30m length, inner diameter 0.25mm, 0.25mm film thickness.
- Temperature program: 110°C to 280°C at a rate of 10°C/min.
- Injector temperature 250°C.
- Injection volume: 2 μl.
- Carrier gas: helium at a constant flow rate of 1 ml/min.

**Results:**

**Phytochemical and antimicrobial result of pumpkin fruit:**

Alkaloids and glycosides, coumarins and flavonoids were found to be present in the ethanolic extract of pumpkin fruit.

Ethanol extract of pumpkin fruit has a high activity against selected gram positives (*Staphylococcus aureus* and *Bacillus subtilis*) and gram negatives bacteria (*Pseudomonas aeruginosa* and *Escherichia coli*).

**GC-MS Analysis Result of Pumpkin Fruit:**

This investigation was carried out to determine the possible chemical components from pumpkin fruit by GC-MS. The results pertaining to the GC-MS analysis of pumpkin fruit were given in table and figure bellow:
Table 1: The Name, Formula and Structures of phyto-chemical components detected in Pumpkin Fruit by GC-MS:

<table>
<thead>
<tr>
<th>№</th>
<th>Name</th>
<th>Formula</th>
<th>Structure</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Vit E</td>
<td>C_{29}H_{50}O_{2}</td>
<td><img src="image" alt="Vitamin E structure" /></td>
</tr>
<tr>
<td>2</td>
<td>5-methyl-5a,6,8,9,10,11,11a,12-octahydro-5H-6,10,11,12a-dimethanoindol[3,2-b]quinolizine-8,13-diol</td>
<td>C_{26}H_{26}N_{2}O_{2}</td>
<td><img src="image" alt="Compound 2 structure" /></td>
</tr>
<tr>
<td>3</td>
<td>1H-imidazole, 2-ethyl-4-methyl</td>
<td>C_{6}H_{10}N_{2}</td>
<td><img src="image" alt="Compound 3 structure" /></td>
</tr>
<tr>
<td>4</td>
<td>Methoxyphenamine</td>
<td>C_{11}H_{17}NO</td>
<td><img src="image" alt="Compound 4 structure" /></td>
</tr>
<tr>
<td>5</td>
<td>1,2-dihydroxyethyl-3,4-dihydroxyfuran-2(5H)-one</td>
<td>C_{6}H_{8}O_{6}</td>
<td><img src="image" alt="Compound 5 structure" /></td>
</tr>
<tr>
<td>6</td>
<td>5-[1-hydroxy-2-(isopropylamino)ethyl]benzene-1,3-diol</td>
<td>C_{11}H_{17}NO_{3}</td>
<td><img src="image" alt="Compound 6 structure" /></td>
</tr>
<tr>
<td>7</td>
<td>2-Amino-9-[3,4-dihydroxy-5-(hydroxymethyl)oxolan-2-yl]-3 H-purine-6-</td>
<td>C_{10}H_{13}N_{5}O_{5}</td>
<td><img src="image" alt="Compound 7 structure" /></td>
</tr>
<tr>
<td>8</td>
<td>4,4′-methylenedis(tetrahydro-1,2H,4-thiadiazine)1,1′,1′-tetraoxide</td>
<td>C_{7}H_{16}N_{4}O_{4}S_{2}</td>
<td><img src="image" alt="Compound 8 structure" /></td>
</tr>
<tr>
<td>9</td>
<td>3,7,11-trimethyl-2,6,10-dodecanetriol</td>
<td>C_{15}H_{26}O</td>
<td><img src="image" alt="Compound 9 structure" /></td>
</tr>
<tr>
<td>10</td>
<td>Tetradecanoic acid</td>
<td>C_{14}H_{28}O_{2}</td>
<td><img src="image" alt="Compound 10 structure" /></td>
</tr>
<tr>
<td>No.</td>
<td>Chemical Name</td>
<td>Chemical Formula</td>
<td>Image</td>
</tr>
<tr>
<td>-----</td>
<td>-------------------------------------------------------------------------------</td>
<td>-----------------</td>
<td>-------------</td>
</tr>
<tr>
<td>11</td>
<td>Squalene</td>
<td>C_{30}H_{50}</td>
<td><img src="1" alt="Image" /></td>
</tr>
<tr>
<td>12</td>
<td>3,7,11,15-tetramethyl-2-hexadecan-1-ol</td>
<td>C_{26}H_{40}O</td>
<td><img src="2" alt="Image" /></td>
</tr>
<tr>
<td>13</td>
<td>1H-Purine-2, 6-dione, 3,7-dihydro-1,3-dimethyl-</td>
<td>C_{7}H_{8}N_{4}O_{2}</td>
<td><img src="3" alt="Image" /></td>
</tr>
<tr>
<td>14</td>
<td>1-β-D-ribofuranosyl-1H-1,2,4trazole-3-carboxamide</td>
<td>C_{8}H_{12}N_{4}O_{5}</td>
<td><img src="4" alt="Image" /></td>
</tr>
<tr>
<td>15</td>
<td>6,7-dimethoxy-3-[(5R)-4-methoxy-6-methyl-7,8-dihydro-5H-[1,3]dioxolo[4.5-g]isoquinolin-5-yl]-3H-2-benzofuran-1-one</td>
<td>C_{22}H_{23}NO_{7}</td>
<td><img src="5" alt="Image" /></td>
</tr>
<tr>
<td>16</td>
<td>1-(4-tert-butylphenyl)-4-[4-[hydroxyl(diphenyl)methyl]piperidin-1-yl]butan-1-ol</td>
<td>C_{32}H_{41}NO_{2}</td>
<td><img src="6" alt="Image" /></td>
</tr>
<tr>
<td>17</td>
<td>2-phenylethyl hydrazine</td>
<td>C_{8}H_{12}N_{2}</td>
<td><img src="7" alt="Image" /></td>
</tr>
<tr>
<td>18</td>
<td>2,6-di(propan-2-yl)phenyl</td>
<td>C_{12}H_{18}O</td>
<td><img src="8" alt="Image" /></td>
</tr>
<tr>
<td>19</td>
<td>4-[3-(5H-Dibenz [b,f] azepin-5-yl)-1-piperazinethanol</td>
<td>C_{23}H_{29}N_{3}O</td>
<td><img src="9" alt="Image" /></td>
</tr>
<tr>
<td>20</td>
<td>5-fluoro-1-[4-hydroxy-5-(hydroxymethyl)tetrahydrofuran-2-yl]-1H-pyrimidine-2,4-dione</td>
<td>C_{9}H_{11}F N_{2}O_{5}</td>
<td><img src="10" alt="Image" /></td>
</tr>
<tr>
<td>21</td>
<td>4-chloro-N-(2-methyl-2,3-dihydroindol-1-yl)-3-sulfamoyl-benzamide</td>
<td>C_{16}H_{16}Cl N_{3}O_{3}S</td>
<td><img src="11" alt="Image" /></td>
</tr>
</tbody>
</table>
Discussion:

Miracles from Quran represent a real challenge in discovering of chemical ingredients and its relationship with remedies. Selection of pumpkin within billion of plants did not come by chance but it was an indirect message from God to search in-depth in the mysteries and secrets of this great plant and through this work, twenty two natural chemical compounds have been detected in...
the investigated part of pumpkin (fruit), they need more efforts and deep analysis and interpretation to elucidate them in the plant. Through the presence of twenty two of natural chemical compounds in pumpkin fruit can clearly see the extent of role of pumpkin fruit in the therapy.

**Activity of the identified phyto-components in pumpkin fruit:**

**Vit. E:**

The primary function of vitamin E is antioxidant (Ritter et al., 2008).

**5-methyl-5a,6,8,9,10,11a,12-octahydro-5H-6,10,11,12a-dimethanoindol [3,2b]quinolizine-8,13-diol:**

Also known as Ajmaline, it is an indol alkaloid and a highly effective anti-arrhythmic drug. It has been used for the treatment of patients with Wolf-Parkinson-White syndrome with paroxysmal atrial fibrillation (Bahnikova et al.,2002; Bébarová et al.,2005;Batchvarov et al.,2009).

**1H-Imidazole, 2-ethyl-4-methyl- or (Imidazole, 2-ethyl-4 methyl):**

On the basis of various literature surveys Imidazole derivatives shows various pharmacological activities include:

- Anti fungal
- Anti-bacterial activity
- Anti inflammatory activity
- Analgesic activity
- Anti tubercular activity
- Anti depressant activity
- Anti cancer activity
- Anti viral activity
- Antileishmanial activity (Shalini et al., 2010).

**Methoxyphenamine:**

It is a sympathomimetic agent which causes bronchodilatation and is used in commercial preparations as a nasal decongestant (Lau et al., 1990). It is frequently used in alleviating the bronchial asthma and symptoms of Chronic obstructive pulmonary diseases COPD including cough, sputum and asthma. However, there are few studies of its anti-inflammatory effect on COPD (Yue-Hong et al., 2003).

**1,2-dihydroxyethyl-3,4-dihydroxyfuran-2(5H)-one:**

Named as VitC or Ascorbic acid is required for the maintenance of normal connective tissue, as well as for wound healing. Vitamin C facilitates the absorption of dietary iron from the intestine (Champe et al., 2008).
Also known as antioxidants, consumption of diets rich in these compounds is associated with a decreased incidence of some chronic diseases, such as coronary heart disease and certain cancers (Champe et al., 2008).

2-Amino-9-[3,4-dihydroxy-5-(hydroxymethyl)oxolan-2-yl]-3 H-purine-6: (Guanosine), it is similar to acyclovir, most effective against herpes simplex virus (Criage&stitzel, 1997).

4,4’-methylene bis(tetrahydro-1,2H,4-thiadiazine)1,1,1’,1’-tetraoxide:  

Taurolidine (TN) is an antibacterial drug originally synthesised in 1970 with activity against a broad spectrum of microorganisms and has been used as a safe antibiotic to prevent bacterial infection in patients after abdominal surgery (Walters et al., 2007; Aceto et al., 2009; Chromik et al., 2010).

Taurolidine also appears to have immunoregulatory properties, blunting lipopolysaccharide induced tumour necrosis and also reducing adherence of bacteria to human epithelial cells in vitro (Torres-Viera et al., 2000).

3,7,11-trimethyl-2,6,10-dodecanetriene-1-ol:  

Farnesol (FOH) is a naturally available sesquiterpene alcohol and a key intermediate in de novo synthesis of cholesterol in all mammalian cells. It acts to suppress Candida albicans development and this isoprenoid alcohol has been demonstrated to inhibit the growth of some bacteria, such as Staphylococcus aureus and some fungal species and recently reports have shown that it can intensify the effect of antimicrobial agents on Staphylococcus aureus (S. aureus) and Escherichia coli (E. coli) (Jing et al., 2010).

Tetradecanoic acid (Myristic acid):  

Epidemiological and clinical studies have shown that dietary fats containing high levels of saturated fatty acids induce an increase in plasma total and low density lipoprotein (LDL) cholesterol concentrations in humans, among saturated FA (RiouxF et al., 2005).

Squalene:  

Squalene is a triterpene, shows some advantages for the skin as an emollient and antioxidant and for hydration and its antitumor activities. It is also used as a material in topically applied vehicles.
Squalene might be a useful addition to potentiate the effects of some cholesterol-lowering drugs, the primary therapeutic use of squalene currently is an adjunctive therapy in a variety of cancers (Smith TJ, RT al, 2000).

1H-Purine-2, 6-dione, 3,7-dihydro-1,3-dimethyl-:
Theophylline remains one of the most widely prescribed drugs for the treatment of asthma and Chronic Obstruction Pulmonary Disease (COPD) world-wide, also has anti-inflammatory effects (Barnes, 2005; Barnes, 2010).

1-β-D-ribofuranosyl-1H-1,2,4triazole-3-carboxamide:
Ribavirin shows broad-spectrum antiviral activity against a variety of viruses and is used in combination with interferon-α to treat hepatitis C virus infection (Crotty et al., 2000).

6,7-dimethoxy-3-[(5R)-4-methoxy-6-methyl-7,8-dihydro-5H[1,3]dioxolo[4.5-g]isoquinolin-5-yl]-3H-2-benzofuran-1-one:
Noscapine (opioid alkaloid) is an antitussive agent(very safe cough suppressant), acting on central nervous system site (Matchett et al., 2001; Nadendla, 2005). However, unlike codeine and other narcotics, noscapine lacks addictive effect. Some studies have shown anxiolytic effects of noscapine in mice. Noscapine effectively inhibits the progression of various cancer types both in vitro and in vivo (Barken et al., 2008).

1-(4-tert-butylphenyl)-4-[4-hydroxyl(diphenyl)methyl] piperidin-1-yl butan-1-ol:
Terfenadine is an antihistaminic formerly used for the treatment of allergic conditions (Zünkler et al., 2000).

2-phenylethyl hydrazine:
Phenelzine and several other MAOIs, such as isocarboxazide, are structurally related to amphetamine and were synthesized in an attempt to enhance central stimulant properties. (Nadendla, 2005).

2,6-di(propan-2-yl)phenyl:
Propofol is a liquid short-acting general anaesthetic. Also it is an intravenous sedative– hypnotic agent that is commonly administered for sedation of patients in the intensive care unit (ICU) (Barr et al., 2001; Kam & Cardone, 2007).

4-[3-(5H-Dibenz [b,f] azepin-5-yl)-1-piperazinethanol:
Opipramolis a tricyclic compound for therapy of anxious-depressive states and general anxiety disorders. It is clinically used in Germany and a few European countries as a substitute for benzodiazepines for long-term application (Möller et al., 2001; Turhan & Uslu, 2008).

5-fluoro-1-[4-hydroxy-5-(hydroxymethyl)tetrahydrofuran-2-yl]-1H-pyrimidine-2,4-dione:

Floxuridine is a fluorinated pyrimidine that is primarily used for the treatment of metastatic carcinoma of the colon and a variety of cancers for the past 40 years (Ning et al., 2007).

4-chloro-N-(2-methyl-2,3-dihydroindol-1-yl)-3-sulfamoyl-benzamide:

Indapamide is a thiazide-type diuretic, a widely used as antihypertensive agent (Sanam et al., 2012).

Canrenone:

Canrenone, a cardiovascular drug, has been widely used as a nonselective aldosterone receptor antagonist clinically to treat heart failure, high blood pressure, oedema, liver ascites, and other cardiovascular diseases (Da-Ming et al., 2011).

Conclusion:

As a consequence of all these bioactive twenty two compounds and their important biological activities, Allah Almighty had chosen the pumpkin tree for the prophet Yunus (pbuh) because of its benefits and usefulness in the large scale in recovering health and strength, and because of the presence of several beneficial active ingredients. However, isolation of individual phytochemicals and subjecting it to biological activity will definitely give fruitful results.
التوصيات: نوصي بإجراء دراسات متقدمة وضرورة عمل الرنين المغناطيسي وتحليل بالأشعة تحت الحمراء وكذلك لابد من دراسة اكلينيكيه لتوضيح العلاقة بين المكونات الكيميائية وخصائصها الدوائية.

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