Study the effect of Indian premium (*Saussurea costus*) extract and sesame oil on the sensitivity of local *Acinetobacter baumannii* isolates causing infection

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Abstract
The current investigation was intended to assess the viability of sesame oil got from Heet city and Indian premium (*saussurea costus*) affectability testing against *Acinetobacter baumanii* isolates detached from disease destination like urine, sputum, C.S.F, wound swab, and burn swab. The results indicated that sesame oil activated the gentamycin antibiotic activity from 3.08 mm to 14.7500 mm and also the Indian premium made the same thing the activation was from 5.4167 mm to 9.3333 mm. while the addition of sesame oil to Imipenem antibiotic there was inhibition in the activity of Imipenem from 21.33 mm to 20.083mm and when we added the Indian premium there was activation to the Imipenem antibiotic was from 22.1667 mm to 23.4167mm. Finally the results of cefotaxime after adding the sesame oil we noticed that there is decrease in the activity of cefotaxime from 3.500 mm to 1.000 mm and we also noticed that the addition of Indian premium to the antibiotic there was decrease in the activity from 3.9167 mm to 3.33 mm. Therefore ower explanation that there is activation to some antibiotics there is also inhibition to the other by adding different plant extract.

Key Words: *Saussurea costus*, sesame oil , *Acinetobacter baumannii*.

Introduction
*Acinetobacter* is variety Gram-negative with the widest Gammaproteobacterial class position. Species of *Acinetobacter* are not motile and oxidase – negative and usually Nitrate –negative (1) patients are *Acinetobacter* genus a vital wellspring of disease in crippled patients in clinic .specifically species Species of *Acinetobacter baumanii* of the family *Acinetobacter* are rigorously vigorous, non-fermentative ,Gram negative bacilli . *Acinetobacter* is frequently separated from Nosocomial conditions and is mostly used in concentrated examination units, in which normal A.baumanii is both incoherent and pandemic, are the continuous reasons for Nosocomial pneumonia especially late-start ventilatory-related pneumonia. (2) The spread of medication safe microorganism is quite possibly the most genuine dangers to effective treatment of microbial sickness(3).Circumstantially the most recent decade has likewise seen expanding escalated concentrates on separates and organic dynamic mixtures disengaged from plant species utilized for characteristic treatments or home grown medication (4),(5) for more than millennia,regular plants have been viewed as an important wellspring of therapeutic specialists with demonstrated capability of treating irresistible illness and with lesser results contrasted with engineered drug specialists .Sesame has a place with the family-pedaliaceae and variety – *sesamum*. The sort comprises of around 36 species and 19 of which are native to Africa. Sesame plant is accepted to have started from Africa (6). In societal medicines in Africa and Asia, sesame is rumored. All plant parts are precious. However, the decoction of the leaves in south-western Nigeria is used for treating injured and expelled skin, eye and fomentation Mixed warm water leaf is used for washing stimulated mouth films. The two leaves and roots were decoction for chicken box and measles and were used in *Taenia capitis* as a hair cleaner (7). Sesame seed oil, known to the man since the start of its development, has been used as recovering oil and privately burnt as a permanent food in Nigeria notably in southwest and central belt areas. The sesame oil consists of mainly four fatty
acids, while other fatty acids appear in very small amounts. Due to the fact that sesame seeds are rich in UFAs, as well as vitamin and minerals such as calcium, magnesium and phosphorus, its oil has health benefits. Therefore, sesame oil has been used as medicine or for pharmaceutical purposes. Sesame oil contains Vitamin E and several important antioxidants such as sesaminol and sesamolinol that are believed to promote the integrity of body-tissues in the presence of oxidizing compounds. Saussurea costus, a premium known in secret as Kuth in Pakistan and India has the overpowering flavor of the Western Himalayas. The species is confined to a small complice of the Himalayas and develops at a height of 2600-4000 meters on moist slopes. The species has evolved during the 1920's and 30's in Himachal Pradesh and Uttarakhand. The species has long been employed in the area's conventional clinical assessment procedures. S. costus is the most industrially suitable species among the Saussurea kinds. Costus has a powerful, sweet scent of severe flavor and is used as an antiseptic, in bronchial asthma management, in particular vagotonic asthma. Courses of action for this species are also provided an explanation for the correction of various contaminations and circumstances. The oil extracted from the roots is known as Costus Oil, which is utilized for the preparation of high quality perfumes and hair oil. In addition, Costus Oil is predicted to be strong in illness therapy. The Saussura costus constituents are a total 35 aroma compounds representing about 92.81% of the total composition were identified. Aldehyde like (7Z, 10Z, 13Z)-7, 10, 13-hexadecaterinal (25.5%) was found as a major compound including other ketones like dehydrocostus lactone (16.7%), alcohols like elemol (5.84%), γ-costol (1.80%), vulgarol B (3.14%), valerenol (4.20%), and terpinen-4-ol (1.60%), etc. Esters and acids were found to be completely absent in our samples.

Materials and Methods
Sample collection:
The samples of sesame oil collected for the study was from Heet city. It was extracted by different farmers of Heet from the locally grown sesame seeds and the sample of Indian premium (Saussurea costus) was brought from the Herb shop in Al-khaldia like powder form. Sesame is a warm season annual crop. sesame is a drought tolerant but doesn't need irrigation when young farmers know when to pick sesame seeds harvest occurs 90-150 days from planting out the crop must be harvested. When the plant is mature the leaves and stem of sesame plant change from green to yellow to red. The leaves start to drop from the plant. If its planted in early june for example the plant will begin dropping leaves and drying out in early October its not ready to pick through it takes a while for the green to disappear from upper seed capsules this is referred as drying down. After the plant is dried completely the capsule that cotaines the seed will automatically explode and shoot out the seeds and then its harvested then the seeds are squeeze out and will get sesame oil.

We identify the sesame plant as an annual plant growing 50 to 100 cm tall with opposite leaves 4 to 14 cm with an entire margin ,they are lanceolate, to 5 cm broad at the base of the plant, narrowing to just 1 cm broad on the flowering stem.

Collection of isolates:
Isolates from various departments of the Alramadi education hospital were taken and these isolates thered from patients following history, then sampled from a variety of samples, using procedures that were discussed in relation to kinds such as urine.,Burn swab ,Wound swab, sputum and C.S.F . from all specimen only 12 The Acinetobacter baumanni has been discovered by utilizing isolates Vitek 2. System.

Identification:
According to the specifications of the manufacturer, recognizable evidence with the VITEK 2 framework was provided via IDGNB cards. 41 tests, 18 sugar osmosis tests, 18 sugar aging tests, 2 decarboxylase tests, and 3 other tests are included on 64-well plastic ID-GNB cards (for urease, use of malonate, and tryptophane deaminase). The cards with vacuum
device are vaccinated with the suspension of the biological entity of 0.5 McFarland prepared for 18 to 20 hours on Macconkey agar plate (bioMérieux). VITEK 2 peruser inoculator module. Fluorescence is estimated each 15 min, and the aftereffects of recognizable proof are resolved after 3 hr.

**Culture media and inoculums preparation:**
After we identified all the isolates we prepared special media used in study as the following:

1- First media was prepared by solving 1.3g of nutrient broth powder in 90ml of DW and then we add 10 ml Indian premium extract which was prepared by solving 100 g of Indian premium in 400 ml boiled and hot DW and left for 24 h and then we take 10ml of the extract after centrifugation. And then put it in autoclave for sterilization. The method of plant extraction is done by adding 100gm of Indian premium in powdered form to 400 ml of hot boiled water then left for 24 hr. in room temperature and after we centrifuge the hole solution and then that 10ml of the leached solution and added to 90 ml nutrient broth.

2- Second media is prepared by solving 1.3g of nutrient broth powder in 90ml DW and then we add 10 ml of sesame oil which was prepared by diluting 3ml of sesame oil in 7ml of DMSO after diluting the sesame oil it was added to 90ml of nutrient broth and then autoclaved for sterilization.

After we finish the peppermint of the media its left to cool up and inoculated by the bacterial isolates which was identified as Acinetobacter baumannii by vitek 2. The isolates were done by solving a colony from the agar media in 5ml normal saline until we get a suitable Mcfarland which was 0.5 measured by a density meter, we inoculate both of nutrient broth which contains Indian premium and that contains the sesame oil.

They are left in overnight incubation we will see turbidity in the broth because of the bacterial growth.

**Agar diffusion method:**
We use this method in order to see the inhibition zone of the antibiotic to the bacteria and it's done by the following way.

We prepare 100ml of Muller Hinton agar and poured in to Petri dishes and left until it gets solidified. After solidification we put a swab in the broth media that contains growth until if touches the surface of the media and spread it on the surface the Muller Hinton agar until we get a homogenized spread and then we spread three which were Imipenim (IPM), gentamycin (GN) and cefotaxim (CTX) anti biotics that used for study at suitable distances on the Muller Hinton agar and when finish we incubate the Petri dishes for 24 hours at 37 C°.

**Measurement of zone inhibition:**
When the incubation time is finished we will see inhibition zones on the surface of the media caused by the effect of the antibiotic on the bacteria the zone is measured from the edge to the cross edge of the zone.

**Results and Discussion**
In the present study the antimicrobial activity was evaluated against Acinetobacter baumannii species in the first sesame oil added to Gentamycin antibiotic were applied on the bacterial isolate has more effect on the bacterial isolate than the addition of Indian premium as seen in table 1, figure 1.
Table 1: The antimicrobial activity of sesame oil after addition to Gentamycin

<table>
<thead>
<tr>
<th>Medias</th>
<th>N</th>
<th>Mean ± Std. Error</th>
</tr>
</thead>
<tbody>
<tr>
<td>Seasame</td>
<td>12</td>
<td>B 10.83±2.99</td>
</tr>
<tr>
<td>Gent+Sesam</td>
<td>12</td>
<td>B 14.75±2.00</td>
</tr>
<tr>
<td>Gent CN10</td>
<td>12</td>
<td>A 3.08±1.40</td>
</tr>
<tr>
<td>Indian prem</td>
<td>12</td>
<td>AB 6.16±2.27</td>
</tr>
<tr>
<td>Genta+Ind prem</td>
<td>12</td>
<td>B 9.33±2.23</td>
</tr>
<tr>
<td>Genta CN10</td>
<td>12</td>
<td>AB 5.41±1.59</td>
</tr>
<tr>
<td>Total</td>
<td>72</td>
<td>B 8.26±0.96</td>
</tr>
<tr>
<td><strong>P ≥ 0.004</strong></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Figure 1: The antimicrobial activity of sesame after addition to Gentamycin antibiotic

While in table 2, figure 2 we see that there is no antimicrobial activity of IPM after the addition of sesame oil nor did the Indian premium addition with IPM.
Table 2: shows that there is no antimicrobial activation of antibiotic to bacterial isolates after addition to sesame and Indian premium

<table>
<thead>
<tr>
<th>Media</th>
<th>N</th>
<th>Mean ± Std. Error</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sesame oil</td>
<td>12</td>
<td>6.41 ± 1.79</td>
</tr>
<tr>
<td>IPM+Sesame</td>
<td>12</td>
<td>B 20.08 ± 4.20</td>
</tr>
<tr>
<td>IPM</td>
<td>12</td>
<td>B 21.33 ± 3.95</td>
</tr>
<tr>
<td>Indian prem</td>
<td>12</td>
<td>A 2.66 ± 1.13</td>
</tr>
<tr>
<td>prem IPM+Ind</td>
<td>12</td>
<td>B 23.41 ± 5.09</td>
</tr>
<tr>
<td>IPM</td>
<td>12</td>
<td>B 22.16 ± 4.00</td>
</tr>
<tr>
<td>Total</td>
<td>72</td>
<td>16.01 ± 1.73</td>
</tr>
</tbody>
</table>

P=0.000

Figure 2: shows the activation of IMP antibiotic after adding Indian premium

In table three we noticed after we done the experiment that there is no activation of antibiotic activity when we add the Cefotaxim with sesame extract and with Indian premium extract as it’s seen in the table 3 and also in figure 3.
Table 3: shows the inhibition of antibiotic activity after adding Indian premium and sesame oil

<table>
<thead>
<tr>
<th>Medias</th>
<th>N</th>
<th>Mean ± Std. Error</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sesame</td>
<td>12</td>
<td>1.00 ± 0.00</td>
</tr>
<tr>
<td>CTX30 + Ses</td>
<td>12</td>
<td>1.00 ± 0.00</td>
</tr>
<tr>
<td>CTX 30</td>
<td>12</td>
<td>3.50 ±1.68</td>
</tr>
<tr>
<td>Indian prem</td>
<td>12</td>
<td>1.00 ± 0.00</td>
</tr>
<tr>
<td>CTX 30+Ind prem</td>
<td>12</td>
<td>3.91 ± 1.55</td>
</tr>
<tr>
<td>CTX 30</td>
<td>12</td>
<td>3.33 ± 1.59</td>
</tr>
<tr>
<td>Total</td>
<td>72</td>
<td>2.2917 ± 0.47</td>
</tr>
</tbody>
</table>

P ≥ 0.182

Figure 3: shows inhibition activity of antibiotic when adding Indian premium extract and sesame oil

The over use of antibiotic in the treatment of infection and the appearance of multi- drug resistant bacterial strains because of mutations (which means resistant to two or more antibiotics). Which leads to find a research towards the study of anti-microbial agents from natural sources such as sesame oil and Indian premium. Sesame oil showed activation with gentamycin antibiotic and also did the Indian premium. While the addition of sesame oil to imipenem antibiotic did not make activation with antibiotic but when we added indian premium there was an activity with antibiotic. However, the addition of sesame oil and Indian premium to the cefotaxime, there was inhibition in the activity of the cefotaxime.
Conclusion

The current investigation means the counter microbial part of the expansion of sesame oil to Gentamycin and anti-toxin which builds the hindrance movement of the Gentamycin medication to the microorganisms. Sesame oil may be considered as the hotspot for improving the new broad-based antibacterial strategy. These outcomes backing and conviction that the expansion of sesame oil to the anti-infection may have numerous drug jobs. In similar study by Oloma et al. (14) who observed that sesame oil did not show any activity against s.aureus and klebsiella spp.and his result also revealed that the sesame oil were active against fungi but other oil didn’t show any activities in these organism .the results of Oloma didn’t agree with our result in which we find that there was activity of sesame oil on the fungi but not on bacteria. Munir et al. (15) in Nigeria found that methanolic and ethanol extract have broad spectrum antimicrobial effect against all the tested pathogenic micro-organisms except Streptococcus pneumoniae and Staphylococcus aureus respectively, while the aqueous extract exhibited inhibitory activity on S. aureus, S. pneumoniae and Candida albicans. The study by Munir et al., agree with the results that we find that there was activity on bacteria when we use the sesame oil.

The sesame sanani oil and dwaini oil have more activity against A. niger and A. flavus. These results do not agree with Anand et al.,(16) who found antibacterial activity of sesame oil against bacteria. S. mutans and Lactobacillus acidophilus were found to be moderate to sensitive to the sesame oil. This study agrees with the results that we find that sesame was activated against bacteria but the sesame oil was not active on (fungi) and this did not agree with our finding.

Acknowledgement:
I might want to thank the university of Anbar, college of science and Biology departmant for their extraordinary acquired assistance by the postgraduate labs .Additionally thanks reached out to both Mr. Mohamed Aljabri and Mr. Hameed Mahmud for supporting this work.

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دراسة تأثير مستخلص قسط الهندي (Saussurea costus) على حساسية عزلات بكتيريا Acinetobacter baumanii المحلية المسببة للخمج

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الخلاصة

كان الهدف من هذا البحث الحالي هو تقييم جدوى زيت السمسم الذي تم الحصول عليه من مدينة هيت وأيضا اختبار فعاليته مستخلص القسط الهندي ضد عزلات بكتيرية Acinetobacter baumanii المзвولة من حالات مرضية مختلفة مثل البلغم، سائل النخاع ألوشيحو، تسمح الحروق، ومن عينات الإدرار، أشارت النتائج إلى أن زيت السمسم نشط فعاليه المضاد الحيوي جنتمايسين حيث أزدادت الفعالية من 3.08 ملم إلى 14.7500 ملم. وإن القسط الهندي أيضا زاد فعاليه الجنتامايسين من 5.4167 ملم إلى 9.3333 ملم. وأيضا لاحظنا في هذه التجربة انه أضافه زيت السمسم إلى المضاد الحيوي اميبينم أنه هناك تثبيط لفعالية المضاد حيث انخفضت الفعالية من 21.33 ملم إلى 20.083 ملم وعندما تم أضافه القسط الهندي إلى الاميبينم كان هناك تنشيط في الفعالية حيث ازدادت زيادة طفيفة من 22.1667 ملم إلى 23.4167 ملم. وأخيرا أظهرت نتائج أضافه زيت السمسم وقسط الهندي إلى المضاد الحيوي سيفوكاتسيم حيث حدد هناك تثبيط لفعالية المضاد عند أضافه زيت السمسم حيث انخفضت الفعالية من 3.500 ملم إلى 1.000 ملم. أيضا حدد تثبيط للفعالية عند أضافه القسط الهندي حيث انخفضت الفعالية من 3.9167 ملم إلى 3.333 ملم. لذلك يمكن تفسير أن هناك تثبيط لبعض المضادات المستخدمة وقابلية تثبيط بعض المضادات عند أضافه المستخلص النباتي.

الكلمات المفتاحية: القسط الهندي ، زيت السمسم ، Acinetobacter baumanii.